EFFECTS OF DRIED DISTILLERS GRAINS WITH SOLUBLES (DDGS) FEEDING STRATEGIES ON GROWTH PERFORMANCE, NUTRIENT INTAKE, BODY COMPOSITION, AND LEAN AND FAT QUALITY OF IMMUNOLOGICALLY CASTRATED PIGS HARVESTED AT 5, 7, or 9 WEEKS AFTER THE SECOND IMPROVEST® DOSE

A DISSERTATION

SUBMITTED TO THE FACULTY OF THE GRADUATE SCHOOL

OF THE UNIVERSITY OF MINNESOTA

BY

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IN PARTIAL FULFILLMENT OF THE REQUIREMENTS

FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY

Gerald C. Shurson, Adviser

December 2014

Acknowledgements

This thesis could not have been completed without the support and help of many people. My love for agriculture, food-producing animals, and pigs is only possible due to the enormous gift and blessing of growing up on a hog farm, where my early days included the stroller view of the farrowing barn and paint-stick wall art in the finishing barn. My parent's dedication and perseverance through many challenges I've experienced are a gift, and provided me with perspective and instilled my core values. In addition, I could not have accomplished this without the left over meals from my mother and pep-talks from my father, who's first-hand experience of the Ph.D. process was invaluable and is an experience that few fathers anddaughters can relate.

To my committee members, Dr. Jerry Shurson, Dr. Lee Johnston, Dr. Bill Dayton, Dr. Ryan Cox, and Dr. Marnie Mellencamp, thank-you for taking me on as graduate student, and providing opportunities to allow me to develop into a more well-rounded professional, and for challenging me outside of my academic comfort zone. I also appreciate your encouragement of my involvement in the pork industry. To Dr. Pedro Urriola, my sincerest thanks for sharing your knowledge, experiences, brainstorming sessions, the many two-minute conversations that turned into 60+ minute conversations about swine nutrition, pigs, research, and career. Your addition to the swine nutrition group has been invaluable and I appreciate your guidance, especially through the preliminary exam process. I thank Dr. William Dayton, Dr. Marcia Hathaway, and Dr. Mike White in the Growth Biology Laboratory for teaching me molecular biology techniques and providing an environment where I was comfortable asking countless questions. To Dr. Ryan Cox, I thoroughly appreciate staying connected and involved with

the meat science crew, and I appreciate the assistance from Mr. Pete Nelson, Mr. Tristan McNamara, Mr. Dallas Dornink, Dr. Kaitlyn Compart, and Mr. Justin Johnston and all undergraduate meat lab workers, and being so accommodating and patient with this intensive project.

To my fellow swine group graduate students past and present, thank you for your generosity of time and willingness to help on such an extensive research project. This research would not have been possible without you. Thank you for pitching in and helping when needed, and of equal or greater importance, helping me through the ups and downs of graduate school, celebrating the highs, and lending an ear during the lows. Particularly I would like to thank Dr. Kaitlyn Compart, Ms. Brenda Reiter, Dr. Emily Glunk, Ms. Gail Carpenter, Dr. Devan Paulus Compart, Ms. Elizabeth Bjorklund, Mr. Sam Fessenden, Ms. Amy Hazel, Mr. Justin Johnston, and Mr. Isaac Salfer. Additionally, I would like to thank all of my other friends spread across the country for your support and understanding, especially Ms. Faith Wentzel, Ms. Erika Dyk, and Ms. Andrea Marihart.

Still none of this would have been possible without the hard work of the staff at the West Central Research and Outreach Center. Thank you to all Farm Animal Attendants, Mr. Mark Smith, and Ms. Adrienne Hilbrands for all of your hard work in the daily care of pigs in this project and all of the other research projects. Additionally, I would like to thank Dr. Jon Anderson for his statistical analysis expertise and advice.

Last, but not certainly not least, many thanks to Zoetis, Inc for providing funding for this research and I would like to express my gratitude particularly to Dr. Marnie

Mellencamp and Dr. Christi Calhoun. It has been a pleasure working with you and I truly appreciate your guidance and mentorship as professional women in animal science.

Table of Contents

List of Tables

List of Figures

CHAPTER 1: Introduction and literature review

I. Introduction

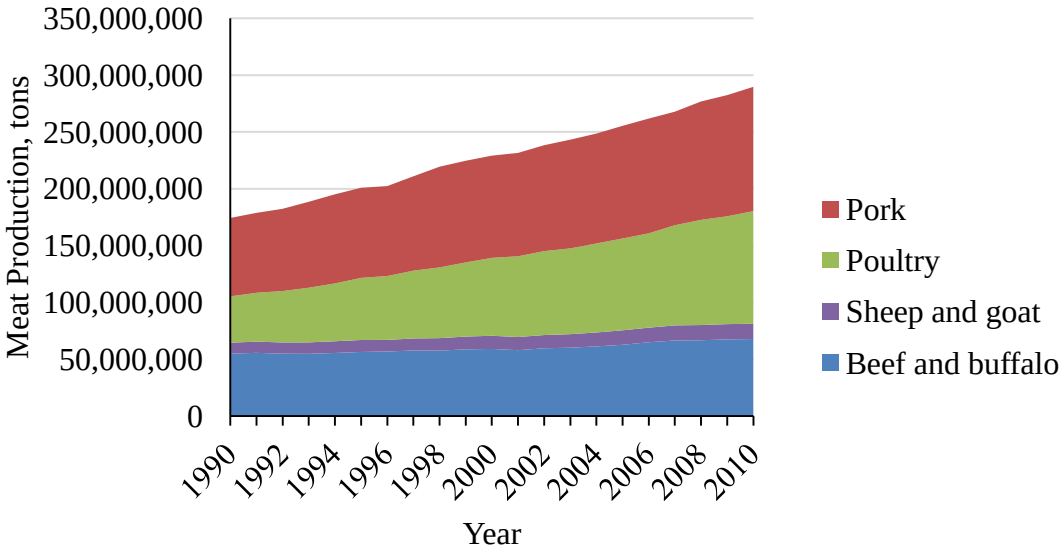
Agriculture will be challenged to feed a growing global population which is expected to increase to 9 to 9.5 million people by the year 2050 (Godfray et al., 2010; Capper, 2011). This population increase will require the use of 60 to 100% more land, water, and energy resources to support human life and produce food (Godfray et al., 2010; Alexandratos and Bruinsma, 2012). As a result, there will be increased competition for resources needed to produce food for humans (Godfray et al., 2010). Population growth is expected to occur in diverging socio-economic classes (FAO, 2013). Greater purchasing power for a growing middle class will increase the number of consumers with the ability to purchase meat (Godfray et al., 2010, Capper, 2011). Meat provides a variety of nutrients vital to the human diet such as protein, long chain n-3 polyunsaturated fatty acid, trace elements, and most B vitamins (De Smet, 2012). These nutrients are more bioavailable from meat than plant-based sources (Godber, 1994), and benefit not only the growing middle class, but also the growing population of undernourished people with insufficient access to calories, protein, and essential nutrients (Godfray et al., 2010; Weldon, 2013). Therefore ensuring the accessibility, availability, and affordability of a nutritious food supply will be necessary for adequate nutrient intake which has human health (Weldon, 2013) and social sustainability implications.

Producing more lean meat with fewer resources can only be accomplished by adopting livestock production practices that focus on managing and conserving finite resources. Therefore, growth of the global food supply will need to come from increased

production and global efficiency of using resources (Godfray et al., 2010).

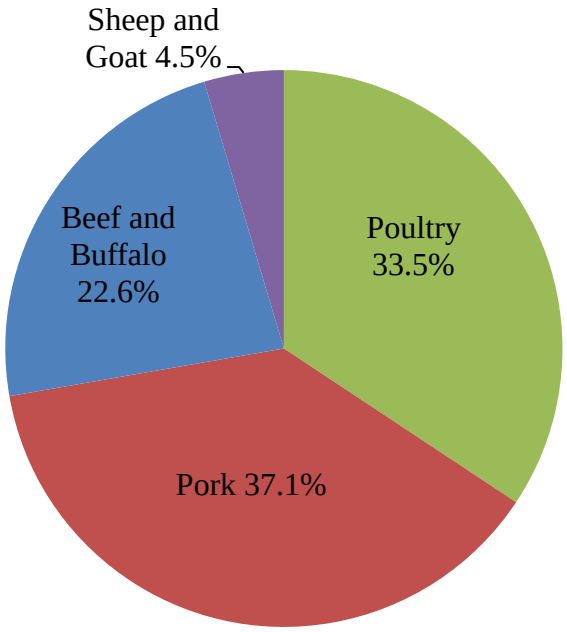
Pork is widely produced and consumed globally, with over 109 million tons of pork produced in 2010 (Figure 1; FAO, 2013). Global per capita pork consumption averages 15.6 kg, which represents 37.1% of total red meat and poultry production (Figure 2; FAO, 2013). The methods used to raise pigs vary widely among countries. In the U.S., pork production practices have drastically changed over the years. A recent life cycle assessment study of the U.S. pork industry characterized several profound changes of pork production practices that have occurred from 1959 to 2009. The results showed a 41% reduction in water use, a 35% reduction in carbon footprint per pound of carcass weight, and a 78% reduction in land use per 1,000 pounds of carcass weight (Boyd and Cady, 2012). The continued growth of the global population underscores the need to continue adopting new technologies and production strategies that improve environmental, economic, and social sustainability.

Figure 1.1. Global meat production (tons) from 1990 to 2010



Adapted from (FAO, 2013).

Figure 1.2. Percentage of total per capita global meat consumption in 2010



Adapted from FAO (2014)

The U.S. is a major contributor to global pork production and consumption. Exports of U.S. pork have increased steadily since 1986, and have rapidly increased since 2004 (USDA-ERS, 2012; Plain, 2013). Becoming a net exporter of pork in 1995 (USDA-ERS, 2012), the U.S. surpassed the EU-27 in total annual pork exports in 2005 (Meyer and Steiner, 2014a), and continues to be the global leader by accounting for 39% of global pork exports in 2008 (USDA-ERS, 2012), and exporting 27% of total U.S. pork production in 2012 (USMEF, 2013). Competition between sources of animal proteins is a result of the cost of substitute animal protein sources (e.g. beef and poultry), as well as cost of pork production among competing countries (Meyer and Steiner, 2014b). Recently, a USDA Agricultural Projections to 2023 report indicated that improvement in production efficiencies will strengthen the global competitiveness of U.S. pork and will

lead to continued increases in U.S. pork exports (USDA, 2014c). In the U.S., feed conversion has improved, and average daily gain has increased from 2007 to 2012, resulting in pigs requiring less feed and fewer days to reach market weight (Stalder, 2012). These improvements in growth performance have been a result of improvements in diet formulation, increased precision of production practices, and use of superior genetics. Consequently, implementation of these technologies has resulted in improved reproductive and lean growth efficiencies which have reduced total cost of production and improved the utilization of resources (Boyd and Cady, 2012).

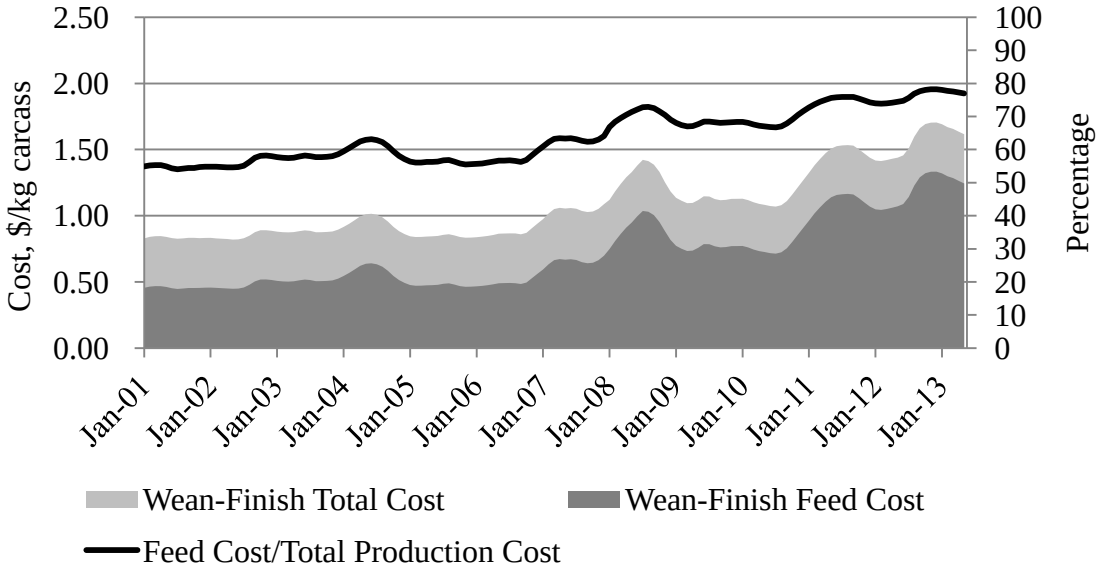
The use of intact male pigs in pork production can be beneficial for reducing the amount of feed consumed, as well as improving lean growth efficiency, energy and nutrient utilization efficiency, and reduce nutrient excretion in manure (Squires, 2011). In the U.S., male pigs have historically been physically castrated to minimize aggressive behaviors between pigs and to prevent unpalatable off-odors of pork from intact male pigs (Sutherland et al., 2010). These limitations need to be overcome in order to capture the energy and nutrient utilization efficiency advantages of intact male pigs. Immunological castration accomplishes both of these objectives and subsequently allows for more lean pork to be produced with fewer resources. Therefore, immunological castration provides environmental and economic benefits that could lead to greater affordability of lean pork. Yet, the use of immunological castration needs to be evaluated with current and evolving feeding practices and genetics used in U.S. commercial pork production systems.

Since 2001, the cost to produce a unit of pork from wean-to-finish pigs has increased more than two-fold from $0.828 to $1.704 per kg of carcass in pigs sold

December 2012 (Figure 3; Schulz, 2014). The general increase in the total cost of pork production has been largely due to the increased cost of feed. In 2001, feed cost accounted for 54% of total production cost and increased to 78.2% for pigs sold in November and December of 2012 (Figure 3; Schulz, 2014). Increased feed cost has been perpetuated by greater demand and price for corn due to U.S. government policies causing increased demand for corn in fuel ethanol production. This has led pork producers to deviate from feeding traditional corn-soybean meal based diets and explore other feed ingredient alternatives (Woyengo et al., 2014). Many alternative feed ingredients have several drawbacks including availability and nutrient consistency. Dried distillers grains with solubles (**DDGS**) is a co-product from the U.S. fuel ethanol industry and has become the most widely available alternative ingredient for corn and soybean meal in swine diets during the last decade. Due to its high relative abundance, relatively low cost, as well as favorable energy, digestible amino acid, and available phosphorus composition, DDGS has become an appealing alternative feed ingredient for use in swine diets (Stein, 2012), with nearly 3 million metric tons used in growing-finishing pig diets in 2008 (USDA-ERS, 2010). Despite the economic and nutritional advantages of feeding DDGS to pigs, there are several challenges. Traditionally, DDGS has contained 10 to 14% corn oil. Corn oil is an rich source of linoleic acid (NRC, 2012) and has led to soft pork fat in carcasses of pigs fed increasing dietary levels of DDGS (Cromwell et al., 2011, Leick et al., 2010, Xu et al., 2010b). Soft pork fat can impose challenges for pork processors by causing poor handling characteristics (Morgan et al., 1994), reduced production yield and quality (Person et al., 2005, Morgan et al., 1994), and decreased shelf-life of bacon (Morgan et al., 1994). Furthermore, the relatively high fiber content in

DDGS often results in reduced carcass dressing percentage (Leick et al., 2010, Xu et al., 2010b, Asmus et al., 2014b), and increased manure output (Xu et al., 2006). The reduction in pork fat quality from feeding linoleic-rich diets is most profound in leaner genotypes such as intact male pigs compared to physical castrates (Wood et al., 2008). These concerns are of particular importance for high fat, further processed products (e.g. bellies and bacon). Therefore, feeding strategies have been developed to optimize dietary DDGS inclusion rates to take advantage of the reduction in diet cost from feeding DDGS, while minimizing the negative effects on pork fat quality of physical castrates and gilt carcasses. However, immunologically castrated pigs have greater nutrient requirements than physically castrated and gilts due to improved lean growth rate and lean gain efficiency. Therefore, there is an industry-wide need to determine the effectiveness of commonly used DDGS feeding strategies for immunologically castrated pigs. Inherently,

Figure 1.3. Feed cost and total cost of U.S. pork production in wean-to-finish pigs sold from 2001 to 2013



Adapted from (Schulz, 2014)

immunologically castrated pigs have less backfat than physically castrated pigs, but increasing the time interval between the second Improvest® dose and harvest increases feed intake and backfat thickness. In theory, the combined production practices of feeding DDGS to immunologically castrated pigs could result in greater sensitivity of immunologically castrated pigs to dietary changes in fatty acid composition, resulting in soft pork fat. These unknowns need to be answered to optimize this combination of production practices and capture the growth advantages of immunologically castrated pigs, minimizing feed cost by adding DDGS to commercial growing-finishing pig diets, and reducing the negative impact of feeding DDGS diets on pork fat quality.

II. Physiological comparison of gilts, physically castrated, and intact male pigs

A. Hormonal profile

The hallmark hormonal differences between intact males, gilts, and physically castrated pigs are the greater levels of serum testosterone in intact male pigs compared to physically castrated pigs and gilts (Clapper et al., 2000), which increases in intact male pigs with advancing age, beginning at 70 days of age (Clapper et al., 2000). Intact male pigs also have greater serum concentrations of estradiol-17β compared to gilts, and greater serum IGF-1 compared to physically castrated pigs and gilts beginning at 84 days of age (Clapper et al., 2000). Estradiol-17β and IGF-1 increase in intact male pigs from 70 to 84 days of age, and continue to increase until 126 days of age (Clapper et al., 2000). More recently, Batorek et al. (2012) reported that serum IGF-1 rapidly increased after 83 days of age, but at 130 days of age, serum IGF-1 was greater in intact male pigs than physical castrates (Batorek et al., 2012). Further increases in serum IGF-1 only occurred in intact male pigs and not physically castrated pigs between 130 to 154 days of age

(Batorek et al., 2012).

Androgens, such as testosterone, elicit an anabolic effect by binding to androgen receptors in the cytoplasm (Squires, 2011). They also have an "anti-catabolic" effect by interfering with glucocorticoid receptors present in high numbers in muscle, limiting the catabolic effect that glucocorticoids have on protein degradation. The androgen-receptor complex enters the nucleus of a cell and stimulates protein synthesis, tissue hypertrophy, and body weight gain (Squires, 2011). Orchietomy and hormonal replacement studies have been conducted in male species to elucidate the mechanism of androgenic effects on growth (Wade and Gray, 1979; Claus and Weiler, 1994). Classically, androgens are known to be necessary to initiate puberty and facilitate development of reproductive organs (Senger, 2003), but they also provide the foundation of physiological feed intake and growth differences between genders (Squires, 2011). At both 147 and 210 days of age, intact male pigs had greater androgen receptor mRNA expression than physically castrated pigs, particularly in the brachialis muscle, but also in the longissimus dorsi and semitendinosus muscles (Yao et al., 2009). Androgen receptors are present in liver, kidneys (Wade and Gray, 1979), as well as muscle (Patience, 2012). The greater circulating testosterone concentrations and androgen receptors suggest that more energy is required by muscle for protein synthesis, thus minimizing excess energy that would be partitioned toward lipogenesis. The increase in protein synthesis and muscle hypertrophy is also supported by the increase in serum IGF-1, which stimulates amino acid and glucose uptake (Clapper et al., 2000). These physiological changes in testosterone, IGF-1, androgen receptor abundance, and muscle hypertrophy are reflected in the improved lean growth rate and efficiency of intact males relative to physically castrated pigs.

There are several different methods used to estimate the proportion of muscle and adipose tissue in live animals. Dual-energy X-ray absorptiometry (**DXA**) is a method that can be used and allows for repetitive body composition assessment throughout the growth period. Results from one study using DXA technology showed that physically castrated and intact male pigs have similar lean deposition from 10 to 122 days of age, but intact male pigs had greater lean deposition from 122 to 150 days of age, resulting in an overall increase in lean deposition from 10 to 150 days of age (Suster et al., 2006). From 66 to 150 days of age, intact male pigs had less fat deposition compared to physically castrated pigs resulting in an overall reduction in fat deposition from 10 to 150 days of age (Suster et al., 2006). The greater fat deposition in physically castrated pigs was also reflected in reduced backfat depth at 150 days of age (Suster et al., 2006). The improvement in protein deposition rate of intact male pigs is 10 to 34% greater than physically castrated pigs as summarized in a review by Millet et al. (2011). The amount of improvement is dependent on body weight range and genetic selection of pigs (including physically castrated pigs) toward greater lean deposition (Millet et al., 2011). Protein deposition at 100 and 150 days of age is 4 and 11% greater, respectively, in intact male pigs than physically castrated pigs (Millet et al., 2011). In the NRC (2012) model, protein deposition is similar between intact male and physically castrated pigs from birth to 60 kg body weight, and increasingly diverges with smaller incremental increases after 90 kg body weight, when maximum protein deposition is achieved in intact male pigs (Figure 1.4; NRC, 2012). Adipose deposition is increasingly greater in physically castrated pigs compared to intact male pigs until 100 kg body weight, when the incremental increase becomes smaller (Figure 1.4; NRC, 2012). These findings are not surprising because

intact male pigs have the inherent advantage of the greater growth rate due to their hormonal regulation of growth. Body composition changes observed between physically castrated and intact male pigs using the DXA scan were also observed in carcasses (Suster et al., 2006). In order to achieve optimal lean growth potential, the energy and amino acid requirements of these tissues must be met (van Milgen et al., 2008).

B. Nutritional requirements to maximize lean and minimize adipose accretion

Pig growth is the balance between energy consuming anabolic and catabolic chemical reactions in the body (Lawrence et al., 2012). Gross energy intake can be supplied from starch, protein, fat, and fiber sources (Patience, 2012). Not all gross energy consumed is available for growth (NRC, 2012). The remaining energy after waste (fecal and urine) products and heat loss can support pig maintenance and growth (NRC, 2012). Energy is partitioned in order of biological importance, with the highest priority given to the brain and central nervous system, followed by bone, skeletal muscle, and adipose tissue (Wray-Cahen, 2001). Energy retention occurs predominantly in the form of protein and lipids (van Milgen and Noblet, 2003; Patience, 2012). In pork production, accretion of lean and adipose tissues are of particular interest since they compromise the largest components of saleable carcass weight (Lawrence et al., 2012). Maximum protein deposition is limited by genetic lean gain potential and the amount of dietary energy and amino acids consumed to meet this potential (van Milgen and Noblet, 2003). Energy intake above maximum protein deposition will be partitioned toward adipose tissue deposition (van Milgen and Noblet, 2003). Deposition of protein requires less energy than adipose tissue deposition due to the lower energy density and greater moisture content of muscle compared to adipose tissue (Table 1.1; Noblet and van Milgen, 2013).

Table 1.1. Metabolizable energy (ME) utilization efficiency and energy requirements for the deposition of adipose and protein in growing-finishing gilts, physically castrated, and intact male pigs

Tissue	ME content of body tissue	ME efficiency to deposit	ME required, dry basis	Wet tissue wt	ME required, wet basis
Adipose	9.4 kcal/g	80%	12.0 kcal/g	1.2 g	7.83 kcal
Protein	5.6 kcal/g	60%	9.5 kcal/g	5.0 g	1.12 kcal

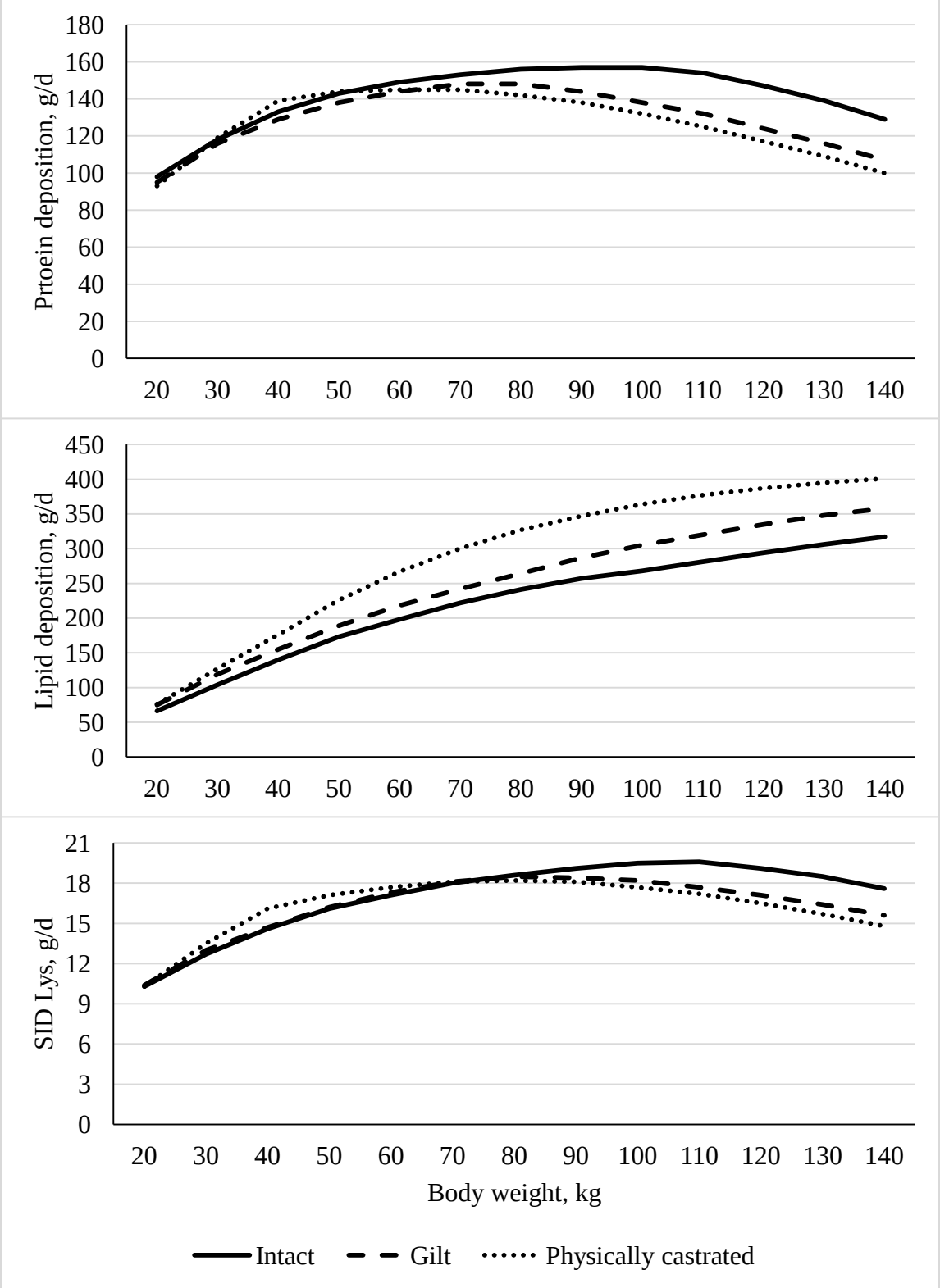
Adapted from (Noblet and van Milgen, 2013).

However, capturing the advantage of greater protein deposition potential can only be achieved by providing adequate amino acids (van Milgen and Noblet, 2003), because they are the functional units for lean growth (NRC, 2012).

In swine diets, lysine is the first limiting amino acid, which means that it is present in the diet in the smallest quantity relative to the pig's requirement compared to all other amino acids (Lewis, 2001). Due to the greater protein deposition in intact male pigs (Figure 1.4), as well as intact males achieving maximum protein deposition at lower energy intake than physical castrates (Quiniou et al., 1996), intact male pigs have a greater standardized ileal digestible lysine requirement than physically castrated pigs and gilts (NRC, 2012, Dunshea et al., 2013).

Traditionally, growth and body composition comparisons of intact male pigs have been compared to physical castrates. However, Dunshea et al. (2013) indicated that there are few such studies involving intact males using contemporary genetic lines with high growth potentials. This has led to comparing the nutritional requirements of intact male pigs to gilts, since it is expected that gilts have a more similar growth pattern to intact male pigs than physical castrates (Dunshea et al., 2013). However, gilts and intact male pigs have diverging nutrient requirements, where gilts have a slightly greater requirement for lysine than physically castrated pigs at 80 kg body weight (Figure 1.4; NRC, 2012), and Dunshea et al. (2013) modeled this divergence to begin at around 70 kg body weight. As a result, protein deposition of gilts is less than intact male pigs, and similar to physical castrates, with gilts having intermediate lipid deposition (Dunshea et al., 1993, Campbell et al., 1989). Still, many studies related to tissue growth curves of immunological castrates use intact males or physically castrated pigs as the reference sex. Nutrient

Figure 1.4. Protein and lipid deposition and required standardized ileal digestible lysine daily intake among intact male, gilts, and physically castrated pigs as modeled by the NRC (2012)



partitioning of energy intake to protein and lipid accretion is regulated by many physiological factors that are under the control of the endocrine system related to gender physiology and growth (Black et al., 2009).

C. Feed intake regulation

Growing-finishing pigs are allowed ad libitum access to feed in U.S. commercial production conditions (Ellis and Augsperger, 2001). Pigs consume feed voluntarily at an amount equal to their energy needs. As a result, pigs fed low energy density diets will consume more feed than pigs fed high energy density diets (Ellis and Augsperger, 2001). Regulation of feed intake and energy metabolism converges at the hypothalamus where orexigenic and anorexigenic signals relay the overall energy status of the body through the arcuate nucleus of the hypothalamus, known as the appetite control center (Squires, 2011). While there are many regulatory signals of appetite, the two opposing primary regulators are adenosine monophosphate-activated protein kinase (**AMPK**) and mammalian target of rapamycin (**mTOR**; Black et al., 2009). When low metabolic energy status is sensed by the elevation of AMP:ATP, AMPK is activated to conserve energy consuming processes (Black et al., 2009). This also inhibits the hypothalamic mTOR, and the mTOR pathway is controlled by leptin and insulin (Black et al., 2009). Leptin is an anorexigenic hormone secreted by adipocytes, and circulating leptin concentrations are directly proportional to body adipose tissue mass, which serves as an energy sensor (Barb et al., 2001). When blood leptin concentration is high, energy status of the body is elevated resulting in reduced feed intake (Barb et al., 2001). A recent study showed that serum leptin concentration did not change from 83 to 154 days of age in intact male pigs, but in physically castrated pigs, serum leptin increased from 130 to 154

days of age, so that by 154 days of age, intact male pigs had lower serum leptin concentration compared to physical castrates (Batorek et al., 2012). The reduced proportion of adipose tissue accretion in intact males relative to physically castrated pigs should result in reduced serum leptin concentrations and increased feed intake. Therefore, these relationships do not explain the reduction in feed intake typically observed in intact male pigs because it is contrary to the convention that increasing backfat results in increasing serum leptin concentration, and thus reduced feed intake. Unfortunately, little is known about the interaction of leptin and androgens in intact male pigs. It may be possible that testosterone, androgen receptors, or an interaction between the two, may play a role in modulating the correlation between leptin, body adipose tissue mass, and feed intake of intact male pigs. One unique characteristic of intact male pigs, which may play a role in modulating the relationship between feed intake and leptin, is the profound production of estradiol-17 β. Estradiol-17 β decreases feed intake (Wade and Zucker, 1970), and intact male pigs have more than 5 times greater circulating estradiol-17 β concentrations than gilts (Clapper et al., 2000). Additional possible modulators of the uncoupling of leptin and feed intake in intact male pigs may be related to reduced feed intake when housed in groups compared to those housed individually (Pauly et al., 2009). Dunshea et al. (2013) summarized that individually housed intact male pigs have similar feed intake compared to gilts. This indicates that hormonal induced behaviors of intact males may be abolishing other known hormonal controls of feed intake typically observed in physical castrates and gilts. Regardless, the major benefits of using intact male pigs in commercial pork production systems are decreased feed intake and greater

lean growth efficiency (Bradford and Mellencamp, 2013). Capturing these advantages would improve overall sustainability of pork production.

III. Limitations of growing intact male pigs in U.S. pork production systems

Capturing the growth performance and carcass composition advantages of intact male pigs in pork production systems is limited due to the presence of boar taint in pork and aggressive boar behaviors (Dunshea et al., 2001). To overcome these drawbacks, intact male pigs are typically castrated within the first two weeks of life in the U.S. (McGlone et al., 1993). Boar taint is detected by some consumers as an unpalatable off odor in cooked pork (Font-i-Furnols, 2012). Aggressive behavior exhibited by intact male pigs compromises animal welfare, results in unnecessary energy expenditure that reduces growth performance potential, and may compromise the safety of animal caretakers. However, the inherent growth performance advantages of decreased feed intake, reduced carcass fat, and improved lean gain efficiency from raising intact male pigs are lost due to physical castration. Physical castration is also being discussed in the U.S. and the EU as a management practice which compromises animal welfare when it is performed without anesthetics or analgesics (Sutherland et al., 2010). Freedom from pain and distress is one of the five freedoms of animal welfare (FAWC, 2009). Physical castration is performed to minimize distress caused by aggressive behaviors that occur among intact male pigs (Guay et al., 2013), however, physical castration also causes acute distress (Sutherland et al., 2010). Use of immunological castration alleviates the practice of physical castration and minimizes aggressive behaviors exhibited by intact male pigs with advancing age. Use of immunological castration also allows capturing the growth advantages of intact male pigs, while mitigating boar taint in pork products. Currently, several alternative

methods to physical castration exist to alleviate the occurrence of boar taint and include: marketing pigs at a lighter body weight, genetic selection against the boar taint causing compounds (i.e. androstenone and skatole), dietary strategies, and changes in the housing environment. However, each of these alternatives has limitations.

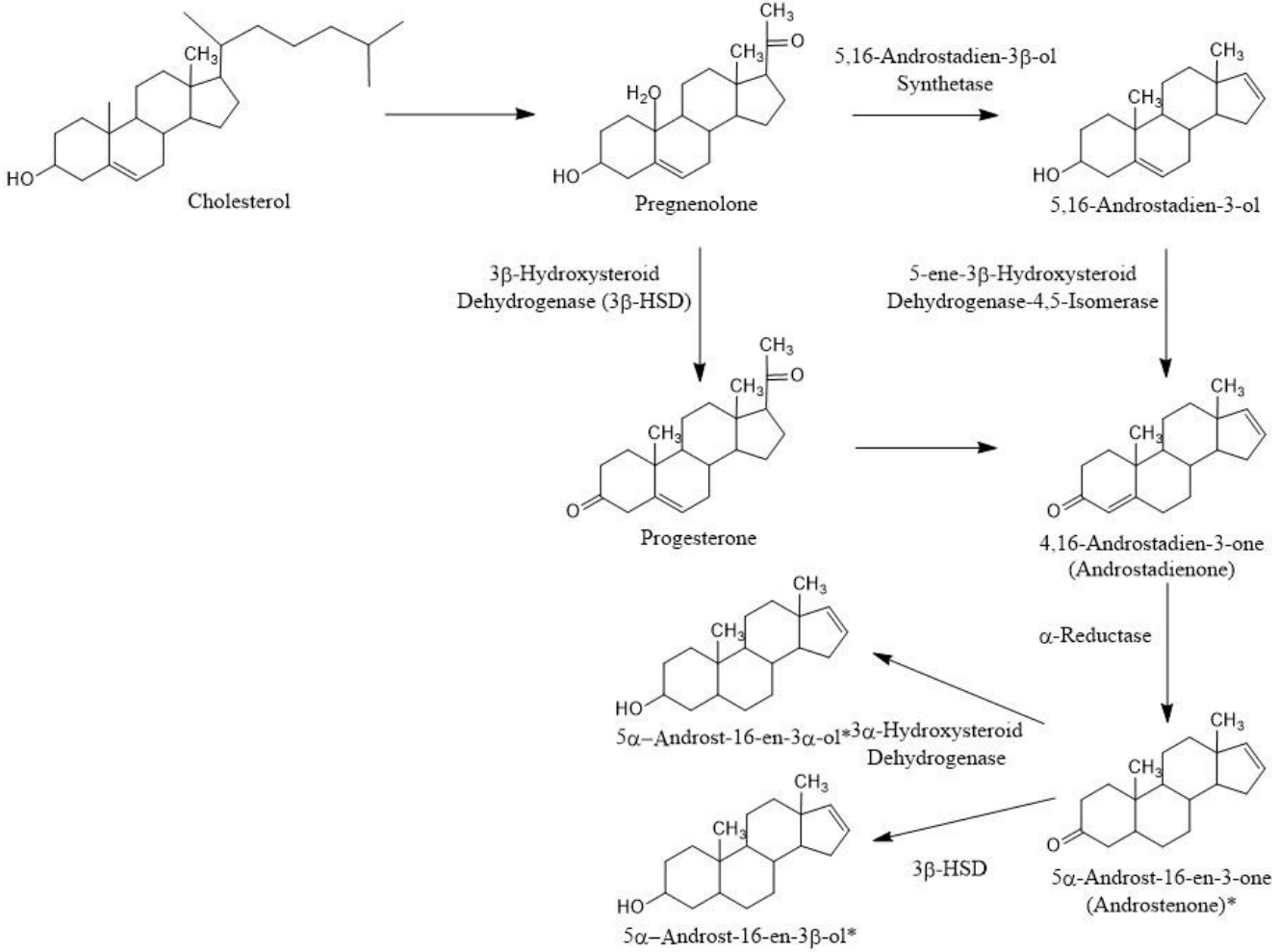
A. Boar taint

1. The role of androstenone and skatole

Boar taint occurs due to the production and accumulation of 5α-androst-16-en-3-one (androstenone; Figure 1.5) and indoles, specifically 3-methylindole (skatole). Androstenone is synthesized from cholesterol as part of the steroidogenic pathway in the testis (Brooks and Pearson, 1986, Bonneau, 1982, Squires, 2011), and skatole is produced from microbial degradation of tryptophan in the large intestine (Squires, 2011, Wesoly and Weiler, 2012). These compounds create an off-odor known as 'boar taint' and result in a negative eating experience for some consumers (Font-i-Furnols, 2012). Not all humans are sensitive or can detect boar taint, but for those who can, it causes a profound, unpalatable aroma. In fact, the Meat Hygiene Regulation 842/2004/EC states that meat with a "pronounced sexual odor" is not fit for human consumption (Whittington et al., 2011). Since these compounds are lipophilic, they accumulate predominately in pork fat and the aroma is most noticeable when pork products are heated.

Androstenone was first isolated from boar testes by Prelog and Ruzicka in 1944, but its association with pork fat and its intense urine-like odor was not established until

Figure 1.5. Biosynthetic pathway of androstenone in the boar testes



Adapted from Brooks and Pearson, 1986); Squires, 2011); Bonneau, 1982)

* can occur in the salivary glands

1968 (Brooks and Pearson, 1986). While there are several C19 Δ16 steroids that are volatile and produce odors, androstenone has the greatest presence in adipose tissue and produces the most intense odor, while other related compounds are more concentrated in the salivary glands (Bonneau, 1982). Production of androstenone originates from the hypothalamus of the brain, which contains clusters of nerve cells called hypothalamic nuclei, that produce neurohormones and releasing hormones that elicit a neuroendocrine reflex (Senger, 2003). Initiating this cascade is the releasing hormone, gonadotropin releasing factor (**GnRF**), which is sometimes referred to as gonadotropin releasing hormone (**GnRH**; Senger, 2003). The axons of the nerve clusters extend to the stalk of the pituitary and meet with the hypothalamic hypophyseal portal system to transport GnRF to the anterior pituitary where follicle stimulating hormone (**FSH**) and luteinizing hormone (**LH**) are released into circulation (Senger, 2003). Follicle stimulating hormone targets the sertoli cells, and LH targets the Leydig cells of the testes where testosterone is synthesized from progesterone and converted to dihydrotestostone, estradiol, and other androgens (Figure 5; Senger, 2003). Androstenone production by the Leydig cells of the testes follows a similar pattern to that of testosterone where production increases rapidly at the onset of puberty (Bonneau, 2006). Androstenone is released into the spermatic vein and systemic circulation (Bonneau, 1982), is primarily deposited in the salivary glands (Bonneau, 1982), and subsequently released through the vomeral nasal passage as a pheromone to stimulate a lordosis response when gilts and sows are in estrus (Senger, 2003). Since androstenone is lipophilic, it also accumulates in adipose tissues resulting in boar taint (Senger, 2003). Production of androstenone by the ovary and adrenal cortex

have also been suggested as a mechanism for low levels of plasma androstenone that have been reported in gilts and physical castrates (Zamaratskaia and Squires, 2009).

In intact male pigs, plasma testosterone and androstenone initially increase from 7 to 14 days of age before peaking, and then decrease after 21 days of age (Sinclair et al., 2001b). This occurs when the hypothalamic-pituitary-gonadal axis is activated (Zamaratskaia and Squires, 2009). It has been hypothesized that adipose androstenone exceeding 4 μg/g at 7 days of age is due to the low amount of adipose tissue in young piglets and the inability to disperse androstenone among fat tissues (Sinclair et al., 2001b). This initial rise of testosterone and androstenone does not influence adipose androstenone concentration at market weight (Sinclair et al., 2001a). Plasma testosterone and androstenone concentrations remain relatively low until 140 days of age when they rapidly increase to 12 ng/ml by 164 days of age (Sinclair et al., 2001b). In most cases, plasma and adipose androstenone concentrations are highly correlated in market weight pigs (Sinclair et al., 2001a; Zamaratskaia et al., 2004; Babol et al., 1999), but the degree of physiological maturity also plays a role. Backfat thickness at market weight is negatively correlated with adipose androstenone concentration, and break-point analysis showed that pigs with less than 14 mm of backfat had increasingly greater plasma testosterone, androstenone, and adipose androstenone than pigs with more than 14 mm of backfat (Sinclair et al., 2001a). It appears that androstenone and skatole preferentially accumulate in adipose tissue with saturated and monosaturated fatty acids, particularly palmitic, stearic, and oleic acids (Mörlein and Tholen, 2015). This relationship has also been demonstrated by comparing Pietrain and Large White breeds (Aluwé et al., 2011), and belly and neck fat (Weiler et al., 1995), where lower androstenone and skatole

concentrations along with greater polyunsaturated fatty acid concentrations occurred in the Pietrain breed and in neck fat (Weiler et al., 1995, Aluwé et al., 2011). Increasing saturated fatty acids and decreasing androstenone and skatole in adipose tissue are opposing goals of U.S. pork production, and could be an additional obstacle for producing intact male pigs since the industry objectives are to minimize boar taint and improve fat firmness (e.g. increase saturated fatty acids).

Accumulation of boar taint occurs due to insufficient metabolism by hepatic 3β-hydroxysteroid dehydrogenase (**3β-HSD**), which when in high abundance, adipose androstenone decreases and androstenone metabolites increase (Doran et al., 2004). Skatole is produced from microbial fermentation of tryptophan derived from intestinal turnover and apoptosis (Claus et al., 2003). Expression of hepatic skatole metabolizing CYP2A and CYP2E1 are greater in physically castrated pigs than intact male pigs (Brunius et al., 2011). The key link between androstenone and skatole is the ability of androstenone to inhibit CYP2E1 (Andresen, 2006; Doran et al., 2002; Doran et al., 2004). This explains the effectiveness of skatole being metabolized in physical castrates resulting in minimal boar taint (Brunius et al., 2011). Controlling androstenone production needs to be a primary target to control boar taint development (Doran et al., 2002). By removing the source of androstenone, hepatic skatole metabolism is allowed to proceed. Alternative strategies would need to achieve similar effectiveness in order to minimize boar taint prevalence when using intact male pigs in pork production systems.

2. Chemical prevalence and sensory detection of boar taint

Sensory thresholds, levels where humans are sensitive to detecting boar taint in adipose tissue, have been established to be 1.0 μg/g for androstenone and 0.25 μg/g for

skatole, with some conservative evaluations using lower sensory thresholds of 0.5 μg/g for androstenone and 0.20 μg/g for skatole (Lundstrom et al., 2009). Consumer sensory evaluation and perception of boar taint has been primarily conducted in European Union countries where intact male pigs are more commonly used in pork production, and thus, the prevalence of boar taint is greater (Font-i-Furnols, 2012; Lundstrom et al., 2009; Babol and Squires, 1995). A multi-national European Union prevalence survey identified differences among countries. Pigs in Spain and Sweden had greater concentrations of adipose androstenone compared to those in Denmark (1.27 and 1.22 vs. 1.04 μg/g, respectively), but all of these levels were above the sensory threshold of 1.0 μg/g of adipose (Walstra et al., 1999). Pigs in Denmark, Spain, and Sweden had higher adipose androstenone concentrations compared to those in the United Kingdom, France, and the Netherlands (0.81 vs. 0.80 vs. 0.79 μg/g, respectively; Walstra et al., 1999).

More recently, a survey was conducted in the U.S. to determine androstenone and skatole prevalence in adipose tissue of physical castrates, gilts, sows, and mature intact male pigs (Prusa et al., 2011). The mean backfat androstenone concentration of physical castrates, gilts, sows, and mature intact male pigs was 0.124, 0.120, 0.100, and 2.363 μg/g, respectively. The concentration of backfat androstenone observed in mature intact male pigs was not surprising and showed that 70.8% of intact male pigs had androstenone concentration greater than 0.5 μg/g, and 55.8% of these boars had concentrations greater than 1.0 μg/g, which exceeded the two commonly used sensory threshold levels. Of greater importance were the androstenone adipose concentrations of physical castrates and gilts. Even at a harvest weight of 69 kg, physically castrated pigs and gilts can have detectable levels of boar taint compounds (Barton-Gade, 1987). In the U.S. pork supply,

2.2% of physical castrates and 1.1% of gilts had greater than 0.50 μg/g backfat androstenone (Prusa et al., 2011). It is likely that cryptorchids could account for the slight prevalence of boar taint in physically castrated pigs observed by Prusa et al., 2011). Sows have been known to produce skatole during estrus (Claus et al., 1994b). In addition, it is possible that some gilts are marketed during estrus which results in a higher probability of them having boar taint because of the heavy market weights used in the U.S. Others have reported boar odor in 65% of mature intact male pigs, 35% of cryptorchids, 5% of physical castrates, 5% of gilts, and 1% of sows (Brooks and Pearson, 1986).

Perceivable boar taint does not necessarily equate to objectionable off-odors (Babol and Squires, 1995). In a review by Babol and Squires (1995), 56% of men and 92% of women were able to detect purified androstenone in solution, and more women than men identified the odor as being objectionable. In fact, in a review by Lundstrom et al. (2009), 8% of humans (16.2% of men vs. 3.3% of women) who were sensitive to detecting androstenone odor actually liked the odor (Lundstrom et al., 2009). An olfactory receptor, ORD7D4, has been linked to the sensitivity and perception intensity of androstenone by humans (Lundstrom et al., 2009). It is also possible that some humans have adapted to the higher prevalence of boar taint and have developed an acceptance of the odor.

The chemical property differences between skatole and androstenone suggest that skatole is more volatile and would be detected first, but that androstenone would have a longer-lasting odor due to its association with fat (Lundstrom et al., 2009). Perception of boar taint in pork products increases with higher concentrations of skatole (Font-i-

Furnols, 2012). Presently in the U.S., there are no analgesics or anesthetics approved for routine on-farm use in swine to minimize pain associated with physical castration. The application of such methods have various obstacles that would need to be overcome for these compounds to be routinely used in commercial pork production systems (Sutherland et al., 2010). In Europe, intact male pigs can be physically castrated without anesthetic except in Ireland, United Kingdom, Portugal, and Spain (Whittington et al., 2011). The United Kingdom and Ireland have banned physical castration, while Norway, Switzerland, and the Netherlands have banned physical castration without the use of anesthetics or analgesics. The European Union is working toward a voluntary ban of physical castration by 2018 (Bradford and Mellencamp, 2013), and some of these countries use alternative production strategies to minimize the occurrence of boar taint. Despite using physical castration to eliminate boar taint, it still exists and physical castration has been criticized for compromising animal welfare. Furthermore, use of physical castration in pork production systems reduces gain and nutritional efficiency, thus requiring more resources to produce pork than using intact male pigs.

IV. Methods to overcome the limitations of intact males in pork production

There are several strategies used to reduce the development of boar taint around the world including harvesting pigs at lower body weights, manipulation of intestinal apoptosis through dietary changes, genetic selection against androstenone, alternative housing environments, and immunological castration. Some of these approaches target androstenone, some focus on skatole, while others aim to target both.

A. Market at lighter body weight

Testosterone and androstenone share similar pathways and are directly associated with puberty attainment. As discussed previously, concentrations of androstenone and skatole change throughout the pig's life. Maturation of boars is known to be body weight dependent. The expression of 3β-HSD is 40% lower in pigs with heavier carcass weights (90 kg) compared to intact male carcasses weighing 70 kg (Nicolau-Solano et al., 2007). In a similar study evaluating six harvest weight groups (body weight range = 90.9 to 113.6 kg; hot carcass weight range from 66.3 to 87.0 kg), 37% of the variation in adipose androstenone concentration was due to differences in body weight. However, in this study, all weight groups had adipose androstenone concentrations greater than 1.0 μg/g, and only pigs harvested with hot carcass weight of 85.2 kg had greater androstenone concentrations than pigs harvested at 75 kg hot carcass weight (Coker et al., 2009). Therefore, pigs with low concentrations of androstenone in adipose tissue were not represented which likely biased the results. Harvesting pigs at lower body weights and before they reach puberty, would reduce androstenone concentrations in adipose tissue. However in the U.S., harvest weights of pigs are much greater than that reported in studies conducted outside of the U.S. Over the last 24 years, U.S. market hog weights have steadily increased at a rate of 0.64 kg/year (Brumm, 2012). More recently, U.S. hot carcass weights have averaged about 100 kg (USDA, 2014b). Therefore, other factors, in addition to market weight, need to be modified in U.S. pork production systems in order to use intact male pigs.

B. Genetics

Androstenone concentration in adipose tissue is a moderately heritable trait (Squires, 2011), which corresponds to its association with testosterone and growth rate, of which are moderately to highly heritable. By selecting for these traits, pigs are inadvertently selected for high levels of steroidogenic hormones, including androstenone, increasing the likelihood of boar taint in pork products. Androstenone concentrations in adipose tissue and salivary glands are lower in maternal breeds such as Landrace and Yorkshire compared with terminal sire breeds such as Hampshire and Duroc (Xue et al., 1996). However, other maternal breeds such as Belgian Landrace and Large White had greater concentrations of androstenone in adipose tissue than Pietrains (Aluwé et al., 2011). Meishan pigs have higher levels of androstenone concentration in backfat, and the rate of β-androstenol (product of androstenone metabolism) is six-fold lower in Meishan pigs compared to Large White pigs, indicating that Meishan pigs have slower androstenone metabolism (Doran et al., 2004). In general, androstenone accumulation appears to be associated with the amount of fat deposition of various breeds, where the Pietrain pigs were the leanest and the Meishan pigs were the fattest. Therefore, selection against androstenone is possible, but selection emphasis on this trait would subsequently result in reduced growth performance and delayed puberty in gilts (Squires, 2011), which is contrary to pork production goals.

C. Diet

Dietary manipulation can be used to reduce boar taint by targeting the reduction of skatole production through modification of the intestinal microflora or substrate available for fermentation. As previously discussed, intestinal turnover appears to be the

primary substrate for fermentation of tryptophan to produce skatole (Claus et al., 1996). Intestinal mucosal turnover is influenced by dietary carbohydrates and protein content. Other factors such as feed form (Jensen, 2006), feed intake behavior (Wesoly and Weiler, 2012), and feed access (Jensen, 2006) contribute to changes in intestinal turnover, but have not been well studied. Results from a feed form study showed that liquid feeding reduced backfat skatole concentrations compared to dry feeding (Andersson et al., 1997), and that fasting pigs for 12 hours reduced adipose skatole concentration (Ambrosen, 1993). However, other feed forms such pellets (Jensen, 2006) and particle size should also be considered because they also alter the microflora in the intestine and cecal nutrient availability.

It is important to consider that any differences in skatole production due to feed form could be dependent on dietary composition and ingredient inclusion. First, the largest proportion of cecal tryptophan is derived from intestinal mucosal turnover (Claus et al., 1996). Feeding energy dense diets, especially when energy is from highly digestible carbohydrates, increases intestinal mucosa turnover, which is likely due to an increase in IGF-1 (Claus et al., 1994b; Claus et al., 1996). Intestinal muscosa turnover is the balance between cellular apoptosis and proliferation (Raab et al., 1998). Insulin-like growth factor-1 mediates the anabolic effect of growth hormone, so when energy intake and growth hormone are elevated, IGF-1 induces intestinal protein synthesis and proliferation (Claus et al., 1996). Not only is IGF-1 production stimulated by the diet, but intact male pigs naturally produce more IGF-1 than physically castrated pigs and gilts (Clapper et al., 2000). Insulin-like growth factor-1 stimulates proliferation of intestinal crypt cells, thus increasing the fermentable substrate available (Claus et al., 1996).

Second, feeding oligosaccharides, resistant starch, or non-starch polysaccharides have been successful dietary strategies for reducing skatole production (Pauly et al., 2011). There are several working hypotheses regarding skatole production and dietary manipulation. The most successful skatole-reducing strategies have resulted in an increase in butyrate production which reduces intestinal apoptosis (Claus et al., 2003). Fructans, such as chicory inulin and raw potato starch, have been the most effective for reducing skatole production, while feeding diets containing sugar beet pulp and soybean hulls have not been effective (Claus et al., 1996). Others studies have targeted a reduction protein fermentation in the hindgut, which have been successful at reducing backfat skatole by either limiting the protein that reaches the large intestine, thus limiting protein fermentation and short chain fatty acid production, or by replacing the protein source with a highly fermentable energy source (Jensen et al., 1995). Increased microbial activity, as measured by ATP production, results in the production of more short chain fatty acids and lowered digesta pH (Jensen et al., 1995). A review by Wesoly and Weiler (2012) discussed other methods of altering the microflora population to reduce skatole production include the addition of antibiotics, organic acids, essential oils from herbs and spices, and tannins.

Increased intestinal transit time increases skatole concentrations in adipose tissue (Wesoly and Weiler, 2012). It is believed that high fiber diets which increase water-holding capacity, dilute the skatole concentration and decrease intestinal transit time to reduce the likelihood of skatole absorption and contact time with intestinal wall (Jensen et al., 1995; Jensen, 2006). Not only can dietary strategies alter skatole production, but the housing conditions or the rearing environment of pigs can influence adipose skatole

concentrations.

D. Environment

Pigs reared in facilities where they have access to manure and bedding have a higher propensity for greater adipose tissue skatole concentrations compared to pigs housed in "clean", non-bedded facilities (Hansen et al., 1994). This finding was determined by altering pig stocking density thus increasing fecal excretion and exposure (Hansen et al., 1994). During summer and winter months, pigs reared in soiled conditions and reduced stocking density, had greater backfat skatole concentrations than pigs reared in “clean” pens with greater stocking density (Hansen et al., 1994). This difference was more profound in summer months (Hansen et al., 1994). These researchers hypothesized that since skatole is a volatile compound, the additional heat during the summer months precipitated greater volatilization of fecal and urine skatole to gaseous form which could have been inhaled through the lungs and absorbed through the skin (Hansen et al., 1994). Modern pork production systems used in the U.S. minimize this mode of skatole accumulation by raising pigs indoors on slatted flooring where no bedding is needed, and most of the manure is contained in storage systems that are inaccessible to pigs.

Intact male pigs can be used in U.S. pork production systems and there are effective management strategies to minimize boar taint in meat products such as reducing market weight, genetic selection, and altering diet composition, and housing management strategies. However, the effectiveness of these strategies in the U.S. might be limited given the current production parameters such as selecting pigs for rapid lean growth and early puberty (e.g. earlier endogenous hormonal production), and marketing at heavier weights compared with market weights used in other countries, which is more likely to

result in greater androstenone production. Immunological castration is one solution to overcome these limitations.

E. Immunological castration

An immunological castration product developed by Zoetis, was first registered in Australia in 1999 under the trade name Improvac (Bradford and Mellencamp, 2013). Since its inception, it has been approved for use in over 60 countries and marketed as Improvest®, Vivax®, and Innosure® (Bradford and Mellencamp, 2013), and for the remainder of this thesis, it will be referred to as Improvest®. In 2011, the U.S. Food and Drug Administration approved the use of Improvest® in the U.S. (Bradford and Mellencamp, 2013) "for the temporary immunological castration (suppression of testicular function) and reduction of boar taint in intact male pigs intended for slaughter" (FDA, 2011c). Use of Improvest® requires a veterinary prescription. The goal of immunological castration is to delay castration until the late finishing phase to capture the growth rate and gain efficiency advantages of boars while allowing time for the metabolism and depletion of previously deposited boar taint compounds in adipose and lean tissues.

Improvest® is a GnRF analog product modified from the endogenous decapeptide and conjugated with a diptheria toxoid protein. This protein is widely used in human vaccines, and for Improvest®, allows for recognition and amplification of the antigen, permitting the pig's immune system to stimulate an immune response generating antibodies against GnRF (Bradford and Mellencamp, 2013). The GnRF antibodies produced from the administration of Improvest® temporarily reduce boar taint in pork by preventing naturally produced GnRF from binding to the GnRF receptors in the

hypothalamus, thus abolishing the cascade of physiological and endocrine changes leading to LH secretion, androstenone synthesis, and boar taint accumulation. Improvest® is administered in two subcutaneous injections at the post-auricular region of the neck by trained technicians. The first dose is administered at least 4 weeks prior to the second dose, and the second dose is administered 3 to 10 weeks before the pigs are harvested. The minimum market time of 3 weeks before harvest fits the previously reported half-life of adipose androstenone of 7 to 19 days when pigs were physically castrated at 90 to 250 kg body weight (Andresen, 2006). Therefore, a minimum marketing period of 3 weeks post-second Improvest® dose has been established to allow sufficient time for any previously deposited boar taint compounds to be metabolized. Two weeks following the second Improvest® dose, a quality assurance inspection is conducted by trained technicians that evaluate each boar individually for aggressive, boar-like mounting behaviors, scrotum color, and testes size (FSIS, 2013). Pigs failing to meet these quality control criteria are administered a third dose of Improvest® and marketed at a minimum of 3 weeks later.

V. Benefits and limitations of immunological castration

A. Physiological changes following the first Improvest® dose

The first dose of Improvest® primes the immune system to generate an antibody response after the second dose is administered (Claus et al., 2008, Zamaratskaia et al., 2008a, Zamaratskaia et al., 2008b). As a result, there are very limited physiological changes that occur after administering the first dose. Compared to intact males, immunologically castrated pigs that only receive the first Improvest® dose have similar average daily feed intake, average daily gain, and feed efficiency (Batorek et al., 2012,

Huber, 2012, Pauly et al., 2009), circulating testosterone (Zamaratskaia et al., 2008a, Dunshea et al., 2001), blood urea nitrogen, and IGF-1 (Huber, 2012). Within 2 weeks after the second Improvest® dose, GnRF antibodies increase rapidly, precipitating a hormonal change from an intact males to resemble physical castrates (Claus et al., 2008, Dunshea et al., 2001,Zamaratskaia et al., 2008a, Zamaratskaia et al., 2008b).

B. Physiological changes following the second Improvest® dose

When the second Improvest® dose is administered to intact male pigs, several physiological changes occur, beginning with changes in plasma concentrations of anabolic hormones. Within the first 14 days following the second dose, LH, testosterone, estradiol 17-β (Claus et al., 2008), and IGF-1 decrease, while blood urea nitrogen increases (Huber, 2012; Claus et al., 2008). Initially at the time of the second dose, 85% of intact male pigs had serum testosterone concentrations greater than 2 ng/mL, but at 2 and 4 weeks following the second dose, 6 to 8% of IC pigs had serum testosterone concentrations greater than 2 ng/mL ("a concentration considered to be biologically significant", respectively; Dunshea et al., 2001). The suppressive effect of Improvest® on testicular function not only affects testosterone production, but also decreases the targeted hormone, androstenone, to decrease boar taint. Circulating concentrations of androstenone and skatole decrease within 14 and 35 days, respectively, after the second Improvest® dose (Claus et al., 2008). Subsequently, these hormonal and metabolic signals alter morphometric size of the testes and accessory glands (Lealiifano et al., 2011, Bonneau, 2010, Batorek et al., 2012), decreases adipose tissue concentrations of skatole and androstenone (Dunshea et al., 2001), as well as alter animal behavior, feed intake, growth rate and gain efficiency (Millet et al., 2011).

Four weeks after the second Improvest® dose, cessation of testicular growth occurs so that at harvest, testicular weight of immunologically castrated pigs is about half of that found in intact male pigs (Dunshea et al., 2001, Pauly et al., 2009). Width and weight, as well as volume and length of testes decrease as the interval between the second Improvest® dose and harvest increases (Lealiifano et al., 2011). Additionally, weight and length of the bulbourethral glands are also markedly reduced, regardless of animal age, when Improvest® is administered and pigs are harvested 4 weeks after the second Improvest® dose (Dunshea et al., 2001, Zamaratskaia et al., 2008a). At 6.5 weeks after the second Improvest® dose, weight of the bulbourethral glands, seminal vesicles, and prostate were dramatically reduced compared to intact male pigs (Batorek et al., 2012). It appears these morphometric decreases of reproductive organs persist until at least 16 to 22 weeks after the second dose of Improvest® (Zamaratskaia et al., 2008b).

Since antibodies decay over time, Improvest® has a temporary effect on the physiological function of the testes, which eventually return to producing testosterone at levels similar to before the second dose of Improvest® was administered (Claus et al., 2008). Mean circulating concentrations of LH, testosterone, and androstenone begin to increase at 11, 13, and 13 weeks following the second Improvest® dose, respectively (Claus et al., 2008). Return to function has been defined when circulating testosterone concentrations exceed 0.5 ng/ml (Claus et al., 2008). The length of time after the second dose of Improvest® to achieve this threshold varies greatly from 74 to 170 days after the second Improvest® dose (Claus et al., 2008).

Several researchers have proposed and attempted to establish the use of

reproductive organ size as a rapid on-line measurement to quantify and segregate carcasses of immunocastrates that may have returned to a functional intact male state (Bonneau and Russeil, 1985; Bonneau, 2010; Dunshea et al., 2001). However, such methods do not seem plausible due to the wide variation of reproductive organ size and mass (Bonneau and Russeil, 1985, Bonneau, 2010, Dunshea et al., 2001, Lealiifano et al., 2011). Lealiifano et al. (2011) assessed colorimetric changes of testes parenchyma in an attempt to establish a real-time measurement to assess pubertal development associated with boar taint. Conceptually, the basis for this assessment utilizes the known peripubertal changes of Leydig cells and intracellular organelle sizes that occur during boar maturity. *In vitro* assessment of testosterone production from Leydig cells showed that there was a high correlation between testosterone production and mitochondrial number and volume (Lunstra et al., 1986). The function of the mitochondria is to produce energy (ATP) in an oxygen-dependent manner, where oxygen is carried by heme-rings resulting in a darker red color, which supports the premise for using colorimetry to detect changes in pubertal development of boars. Additionally, increased LH production causes an increase in testicular blood flow and red blood cells leading to darker parenchyma tissue (Wise et al., 2003).

When using Minolta colorimetry to determine the capacity for testicular function and boar taint accumulation, the parenchyma of intact male pigs was darker and more red compared to parchencyma of immunologically castrated pigs, regardless of whether pigs were harvested at 2 to 6 weeks after the second Improvest® dose (Lealiifano et al., 2011). In order for parchenyma colorimetry to be an applicable tool for packers to use as a screening method, it needs to be evaluated using pigs with greater time intervals after the

second Improvest® dose. It has been suggested that pigs with testicles exceeding 170 g, and having an L* less than 53.0, are indicative of carcasses suspect for boar taint (Lealiifano et al., 2011).

The previously described changes in hormonal profile results in decreased androstenone and skatole concentrations in adipose tissue of immunologically castrated pigs when harvested at 4 weeks after the second Improvest® dose compared to intact male pigs, regardless of age (Dunshea et al., 2001). Adipose tissue androstenone and skatole concentrations exceeded the respective 1.0 µg/g and 0.2 µg/g acceptability threshold in 0% and 3% of immunologically castrated pigs harvested 4 weeks after the second Improvest® dose, respectively, while 49% and 11% of aged-matched intact males exceeded the acceptability thresholds (Dunshea et al., 2001). The percentage of carcasses exceeding 1.0 µg/g adipose androstenone threshold was lower in immunologically castrating pigs harvested 4 weeks after the second Improvest® dose (Dunshea et al., 2001) compared to a recent U.S. prevalence survey identifying 2.8% of physically castrated and gilts exceeding the 1.0 µg/g adipose androstenone threshold (Prusa et al., 2011). Improvest® is highly effective at reducing adipose tissue concentrations of skatole and androstenone.

C. Growth performance

1. Average daily feed intake after the second Improvest® dose

Following the second Improvest® dose, researchers have reported that average daily feed intake is greater in immunologically castrated pigs than intact males and gilts (Table 1.2.; Asmus et al., 2014b; Batorek et al., 2012; Dunshea et al., 2001; Pauly et al., 2009; Puls et al., 2014). Average daily feed intake of immunologically castrated pigs is

either similar to (Asmus et al., 2014b; Batorek et al., 2012; Pauly et al., 2009; Dunshea et al., 2001) or greater (Puls et al., 2014; Asmus et al., 2014b; Dunshea et al., 2001) than physical castrates. The differences in responses across studies could be due to a combination of differences in body weight at harvest and the interval between the second Improvest® dose and harvest. Asmus et al. (2014b) and Dunshea et al. (2001) included two marketing groups of pigs in their studies. In both studies, average daily feed intake (after the second Improvest® dose) of the older age groups was greater in immunologically castrated pigs than in physically castrated pigs, but in the younger age groups, average daily feed intake was similar between immunologically and physical castrates (Dunshea et al., 2001; Asmus et al., 2014b). Moreover, the older pigs evaluated in the Asmus et al. (2014b) study were marketed at a heavier weight (mean = 128.6 kg) and had a longer interval between the second dose and harvest than other studies. In the study by Puls et al. (2014), pigs were much heavier (mean = 131.7 kg) than body weights reported by others.

The changes in average daily feed intake following the second dose of Improvest® have been characterized to not only increase beginning 5 to 6 days after the second Improvest® dose, but also to progressively increase as the time period after the second Improvest® dose increases (Elsbernd et al., 2014). In the extensive evaluation reported by Elsbernd et al. (2014), average daily feed intake was similar to physically castrated pigs by 9 to 10 days after the second Improvest® dose. By 13 to 14 days after the second Improvest® dose, average daily feed intake of immunologically castrated pigs exceeded that of physically castrated pigs (Elsbernd et al., 2014). At 21 and 28 days after the second dose of Improvest®, average daily feed intake of immunologically castrated

pigs was 109% and 113%, respectively of physical castrates (Elsbernd et al., 2014). This could explain the described lack of differences between immunological and physical castrates at older ages.

2. Average daily gain and feed efficiency after the second Improvest® dose

Even though average daily feed intake responses vary during the period between second Improvest® dose and harvest, average daily gain of immunologically castrated pigs is consistently greater than intact males, physical castrates, and gilts during this time period (Table 1.2). Even in studies when average daily feed intake was not different between physically and immunologically castrated pigs (Asmus et al., 2014b; Batorek et al., 2012; Pauly et al., 2009; Dunshea et al., 2001), the improvement in feed efficiency of immunologically castrated pigs compared to physical castrates resulted in greater average daily gain of immunologically castrated pigs. In studies where pigs were marketed at a lighter body weight and younger age, feed efficiency was similar between immunologically castrated and intact male pigs (Batorek et al., 2012; Pauly et al., 2009; Dunshea et al., 2001). This is in contrast to pigs that were marketed 1 to 2 weeks older and more than 10 kg heavier, where immunologically castrated pigs had improved gain efficiency (Puls et al., 2014) and feed efficiency (Dunshea et al., 2001) compared to intact male pigs.

3. Overall growth performance

Growth performance of immunologically castrated pigs, in comparison with other sexes, is more variable when considering the entire growing-finishing period. Overall average daily feed intake of 3 marketing groups in 2 studies (Asmus et al., 2014b; Pauly et al., 2009) was reduced in immunologically castrated pigs compared to physically

Table 1.2. Summary of growth performance of intact male (IM), gilt (G), and physically castrated (PC) pigs compared with pigs immunologically castrated (IC) with Improvest®[1,2]

	Body weight, kg		Harvest, week[3]		Average daily feed intake, kg/d				Average daily gain, g/d				Feed or gain efficiency[4]			
Study	Initial	Harvest	Second Improvest® dose	Age	IM	G	PC	IC	IM	G	PC	IC	IM	G	PC	IC
Between second Improvest® dose and harvest																
Puls et al. (2014)	67.2	131.7	5	25	2.80^{c}	2.90^{c}	3.17^{b}	3.62^{a}	1006^{b}	942^{b}	957^{b}	1179^{a}	0.358^{a}	0.325^{bc}	0.302^{c}	0.326^{b}
Asmus et al. (2014)	24.2	124.7[5]	5	25	X	X	3.10	3.14	X	X	952^{b}	1042^{a}	X	X	0.308^{a}	0.332^{b}
Asmus et al. (2014)	24.2	128.6[5]	7	28	X	X	3.10^{b}	3.22^{a}	X	X	954^{b}	1043^{a}	X	X	0.308^{a}	0.324^{b}
Batorek et al. (2012)	28.4	107.2	5	24	3.11^{a}	X	3.52^{b}	3.46^{b}	956^{a}	X	941^{a}	1131^{b}	3.35^{b}	X	3.84^{a}	3.15^{b}
Pauly et al. (2009)	27.7	107.0	5[6]	23[6]	2.62^{b}	X	3.09^{a}	3.10^{a}	1030^{b}	X	1007^{b}	1136^{a}	2.55^{b}	X	3.08^{a}	2.74^{b}
Zamaratskaia et al. (2008)	27.7	125.1	4	24	NA	X	NA	NA	1107^{a}	X	1090^{a}	1257^{b}	NA	X	NA	NA
Dunshea et al. (2001)	53.9	98.1	4	23	2.44^{a}	X	2.91^{b}	2.81^{b}	786^{a}	X	809^{b}	868^{c}	3.03^{a}	X	3.39^{b}	3.05^{a}
Dunshea et al. (2001)	54.0	117.0	4	26	2.79^{a}	X	3.13^{b}	3.40^{c}	858^{a}	X	847^{a}	1119^{b}	3.30^{b}	X	3.73^{c}	3.10^{a}
Overall																
Puls et al. (2014)	67.2	131.7	5	25	2.68^{b}	2.75^{b}	3.06^{a}	3.11^{a}	1064^{b}	954^{c}	1024^{bc}	1150^{a}	0.397^{a}	0.347^{c}	0.335^{c}	0.371^{b}
Asmus et al. (2014)	24.2	124.7[5]	5	25	X	X	2.30^{a}	2.18^{b}	X	X	915^{b}	932^{a}	X	X	0.398^{b}	0.428^{a}
Asmus et al. (2014)	24.2	128.6[5]	7	28	X	X	2.37^{a}	2.29^{b}	X	X	919^{b}	942^{a}	X	X	0.388^{b}	0.411^{a}
Batorek et al. (2012)	28.4	107.2	5	24	2.46^{a}	X	2.80^{b}	2.68^{b}	914^{a}	X	954^{ab}	1015^{b}	2.71^{b}	X	2.96^{c}	2.64^{ab}
Pauly et al. (2009)	27.7	107.0	5[6]	23[6]	2.06^{c}	X	2.36^{a}	2.22^{b}	883^{x}	X	931^{y}	920^{xy}	2.34^{c}	X	2.54^{a}	2.41^{b}
Zamaratskaia et al. (2008)	27.7	125.1	4	24	NA	NA	NA	NA	971	X	997	1007	NA	NA	NA	NA

[1] Studies that included the interaction between Improvest® and other nutrient partitioning agents (somatropin and ractopamine), did not provide sufficient information to determine the interval between the second Improvest® dose, or did not specify the use of the commercially available Improvest® were excluded from this summary.
[2] Dunshea et al. (2001) only determined growth performance after the second dose of Improvest®.
[3] When age was given in days, age was converted to weeks and rounded up to the whole week.
[4] Depending on the study, some reported efficiency as gain efficiency (values < 1.0) other have reported feed efficiency (values > 2).
[5] Calculated by dividing HCW by (percentage carcass yield divided by 100).
[6] Calculated by the difference between BW at the time of second Improvest® dose and harvest, divided by reported ADG, plus the age at second dose Improvest® dose.
[a,b,c] Within a row of a given variable, means without a common superscript differ ($P \leq 0.05$) as analyzed in respective cited studies.
[x,y] Within a row of a given variable, means without a common superscript differ ($P \leq 0.10$) as analyzed in respective cited studies.
X = Sex not included in the given study.
NA =Unable to determine if the reported means includes growth performance of the entire growing-finishing period or growth performance between second dose and harvest.

castrated pigs. However, 2 other studies (Batorek et al., 2012, Puls et al., 2014) showed similar overall average daily feed intake between immunologically and physically castrated pigs.

Overall average daily gain was greater in immunologically castrated pigs compared to physically castrated pigs only in studies where pigs were older (greater than 25 weeks of age) and had heavier body weights (greater than 124.7 kg) at the time of marketing. Regardless of age, body weight at marketing, or interval between second Improvest® dose and harvest, overall gain or feed efficiency was improved in immunologically castrated pigs compared to physical castrates, but compared to intact male pigs, efficiency was either poorer (Pauly et al., 2009, Puls et al., 2014) or equal to (Batorek et al., 2012) the efficiency of immunologically castrated pigs.

4. Future understanding of feed intake in immunologically castrated pigs

The variable changes in average daily feed intake reported in the scientific literature suggest that the rate of increase and overall response may be a function of age and body weight when the second Improvest® dose is administered. Delaying the second dose of Improvest® may result in a greater or more rapid increase in average daily feed intake than what was observed by Elsbernd et al. (2014). Fortunately, it appears that gain efficiency in immunologically castrated pigs is still improved compared to physically castrated pigs, so the increase in feed intake would result in increased average daily gain. Understanding the dynamics of changing feed intake during the immunological castration process is crucial for determining the appropriate dietary concentrations of energy and nutrients when formulating diets to achieve optimal lean growth, while minimizing nutrient excretion and the cost of lean gain.

D. Environmental impact

Nutrients excreted in swine manure have value when applied to crop land as fertilizer, and serve as a renewable nutrient source for crop production (Kornegay and Verstegen, 2001). However, relative to pork production, nutrients excreted in manure are an economic loss because they were consumed but not used for growth. Furthermore, increased nitrogen and phosphorus content in manure reduces manure application rates to crop land because excess application can compromise soil and water quality (Kornegay and Verstegen, 2001).

Improvement in protein deposition for lean growth increases nitrogen retention and decreases nitrogen excretion in manure because protein contains about 16% nitrogen (NRC, 2012). The benefits of increased protein deposition and nitrogen retention of immunologically castrated compared to physically castrated pigs persist during the first week after the second Improvest® dose, but beginning 9 days after the second Improvest® dose, immunologically and physically castrated pigs had similar nitrogen retention and excretion (Huber, 2012). Maximizing body retention and minimizing excretion of nutrients in manure improves nutrient utilization efficiency, reduces cost of lean growth, and has an environmental benefit. Therefore, appropriate diet formulation of pigs following the second Improvest® dose is necessary in response to changes in feed intake, nitrogen metabolism, and lean growth, and to limit the nutrient cost and environmental impact of overfeeding nutrients. Recently, an environmental life cycle assessment of the impact of Improvest® was conducted. Improvest® was awarded an Environmental Product Declaration for its overall effect of producing more pork and reducing the environmental impact of pork production (De Moraes et al., 2013). This

award exemplifies the substantial benefit of immunological castration on improving nutrient utilization and reducing nutrient excretion in manure.

E. Carcass components

Lean and adipose tissue represent the greatest proportion of total mass in pork carcasses (Lawrence et al., 2012), and are the primary determinants of overall pork carcass value and pork production profitability. Recently, some U.S. pork packers have begun to rely less on using measures of leanness for determining lean premiums and discounts for pork carcasses, and are determining carcass value based on hot carcass weight only. Minimizing fat deposition and improving lean gain efficiency improves carcass value and reduces feed cost because lean gain is less costly than fat gain. Achieving superior lean growth, is not only a consideration for pork producers to improve profit margins, but also has implications on the composition and yield (dressing percentage) of pork carcasses.

1. Carcass dressing percentage

Carcass yield is the proportion of live animal weight that remains in the carcass after exsanguination, dehairing, head removal, and evisceration (Lawrence et al., 2012). The viscera accounts for the greatest proportion of pig body weight at 20 kg of body weight, and this proportion decreases exponentially from 20 to 125 kg body weight (de Lange et al., 2003). Relative growth of the viscera is small during the growing-finishing period compared to lean and adipose tissue (de Lange et al., 2001), but at harvest, visceral mass is the largest non-carcass component removed, and contributes to differences in dressing percentage, or carcass yield, among market pigs (Lawrence et al., 2012). Visceral organs are vital for ingestion, digestion, and metabolism of nutrients, but

account for 50% of whole-body energy expenditure and protein turnover, which cause inefficient use of energy for productive purposes (de Lange et al., 2001). However, visceral organs contribute value when marketed by packers and processors as variety meats and other products.

In 2013, $912 million of variety meats were exported (USMEF, 2014). Exported pork by-products represented a value of $7.66 per head in 2012, which was an increase from $2.23 per head obtained in 2001 (Plain, 2013). Interestingly, it is commonly assumed that visceral organs contribute little to the economic value of market pigs, and that visceral organs and fat tissues are "of low value to the meat processing industry and the consumer" (de Lange et al., 2001). While this may have been true over a decade ago, and is considered to be relatively minor compared to the value of lean tissue, the contribution of these carcass by-products to overall carcass value has increased.

Carcass yield of immunologically castrated pigs is reduced by 1.2 (Pauly et al., 2009) to 3.03 (Yuan et al., 2012) percentage units compared to physically castrated pigs (Asmus et al., 2014b; Boler et al., 2014; Boler et al., 2012; Gispert et al., 2010; Pauly et al., 2009; Boler et al., 2011; Yuan et al., 2012), but carcass yield is similar between immunologically castrated and intact male pigs (Boler et al., 2014; Pauly et al., 2009). Thus, the proportion of hot carcass weight relative to live body weight is lower in immunologically castrated pigs compared to physically castrated pigs. Weight not present in the hot carcass decreases the overall value of the carcass for the producer by reducing hot carcass weight, and thus, increases the cost of production per unit of carcass weight. Immunologically castrated and intact male pigs have an obvious presence of a more developed reproductive tract compared with physically castrated pigs, because physical

removal of the testes early in life results in the absence of testes and less developed accessory glands at harvest (Boler et al., 2014). However, the increased mass of the reproductive tract of intact male and immunologically castrated pigs does not account for all of decrease in carcass yield compared with physically castrated pigs. A detailed evaluation was conducted by (Boler et al., 2014) to identify carcass yield losses among sexes. In that study, carcass yield of immunologically castrated pigs was 1.43 percentage units lower than physically castrated pigs (Boler et al., 2014). The reproductive tract, full intestinal tract, liver, and blood were 0.41, 0.43, 0.13, 0.31 percentage units greater, respectively, in immunologically castrated pigs than physically castrated pigs when expressed as a percentage of live body weight (Boler et al., 2014). Others have also reported that the weight of the liver was greater in immunologically castrated pigs than physically castrated pigs, but similar to intact male pigs (Pauly et al., 2009). The decreased carcass yield caused by greater organ mass is attributed to androgen receptors on organs that are responsive to circulating testosterone, inducing protein synthesis, and thus contributing to body weight gain (Wade and Gray, 1979). From a nutritional perspective, an increase in organ mass, particularly the intestines and liver, result in increased maintenance energy and nutrient requirements (de Lange et al., 2001, NRC, 2012) and thus, increased cost of animal growth.

2. Carcass lean and fat

Decreasing carcass fat satisfies the consumer's preference for meat products with less fat (NPB, 2013). In most cases, backfat thickness of immunologically castrated pigs is reduced (Boler et al., 2012; Dunshea et al., 2001; Pauly et al., 2009; Yuan et al., 2012) or tends (Asmus et al., 2014b) to be reduced compared to physically castrated pigs (Table

1.3). However, Boler et al. (2014) reported that backfat thickness was similar between immunologically and physically castrated pigs. In this study, pigs were marketed at a mean body weight of 135.7 kg, which was greater than market weights of pigs in any other study, but more typical of the current harvest weight of pigs in the U.S. It is important to note, that these pigs were harvested at 5 to 7 weeks after the second dose of Improvest®, but this variable was not considered when interpreting the results of the study.

Loin depth and longissimus muscle area measurements are used to assess the degree of muscling in pork carcasses. The effect of immunological castration on pork carcass muscling appears to be dependent on the measurement method used (Table 1.3). Carcass longissimus muscle area has been reported to be similar between physically and immunologically castrated pigs (Asmus et al., 2014b; Yuan et al., 2012), and among intact males, gilts, physical castrates, and immunologically castrates (Boler et al., 2014), and among intact male, physically castrated, and immunologically castrated pigs (Batorek et al., 2012). However, loin depth was reduced (Boler et al., 2014) or tended (Boler et al., 2012) to be reduced in immunological castrated pigs compared to physical castrates when measured using the Fat-O-Meter.

Backfat thickness and longissimus muscling are combined with hot carcass weight to estimate carcass lean percentage (Table 1.4). The variable responses of backfat thickness and loin muscling in immunologically castrated pigs have resulted in variable estimates of carcass lean percentage. Boler et al. (2012) observed that immunologically castrated pigs tended to have greater carcass lean percentage when using the Fat-O-Meter. However, Batorek et al. (2012) observed no difference, and Zamaratskaia et al.

Table 1.3. Summary of backfat depth and longissimus muscle depth and area among intact male (IM), gilt (G), physically castrated (PC), and intact male (IM) pigs in comparison with immunologically castrated (IC) with Improvest®[1]

	Harvest[2], wk			Backfat depth, mm				Longissimus muscle depth, mm				Longissimus muscle area, cm^2			
Study	BW at harvest, kg	Second Improvest® dose	Age	IM	G	PC	IC	IM	G	PC	IC	IM	G	PC	IC
Asmus et al. (2014)	124.7[5]	5	25	X	X	20.7^{y}	18.5^{x}	X	X	67.8	66.5	X	X	48.6	49.6
Asmus et al. (2014)	128.6[5]	7	28	X	X	24.3^{y}	24.1^{x}	X	X	67.9	67.8	X	X	55.1	54.4
Boler et al. (2014)[3]	135.7	5,6,7	25,26,27	18.2^{a}	21.1^{b}	25.2^{c}	24.3^{c}	-	-	-	-	56.6	58.8	56.1	56.1
Boler et al. (2014)[4]	135.7	5,6,7	25,26,27	23.2^{a}	26.0^{b}	28.9^{c}	28.5^{c}	63.3^{a}	63.7^{a}	66.3^{b}	64.0^{a}	-	-	-	-
Batorek et al. (2012)	107.2	5	24	9.8^{a}	X	14.8^{b}	12.9^{b}	68.7	X	68.4	67.4	46.9	X	45.9	45.6
Boler et al. (2012)[4]	124.5	4	23	X	X	17.7^{b}	16.3^{a}	X	X	63.0^{y}	60.8^{x}	X	X	-	-
Boler et al. (2012)[4]	127.8	6	25	X	X	17.6^{b}	16.3^{a}	X	X	62.6^{y}	61.2^{x}	X	X	-	-
Yuan et al. (2012)	99.2	4	25	X	X	19.1^{b}	13.7^{a}	X	X	-	-	X	X	55.9	51.7
Pauly et al. (2009)	107.0	5[6]	23[4]	17.8^{c}	X	24.9^{a}	19.3^{b}	-	X	-	-	-	X	-	-
Dunshea et al. (2001)	98.1	4	23	11.1^{a}	X	14.4^{b}	11.9^{a}	-	X	-	-	-	X	-	-
Dunshea et al. (2001)	117.0	4	26	12.6^{a}	X	17.1^{c}	15.1^{b}	-	X	-	-	-	X	-	-

[1] Studies that included the interaction between Improvest® and other nutrient partitioning agents (somatropin and ractopamine), did not provide sufficient information to determine the interval between the second Improvest® dose, or did not specify the use of the commercially available Improvest® were excluded from this summary.
[2] When age was given in d, age was converted to wk and rounded up to the whole wk.
[3] Backfat measured by ruler.
[4] Fat-O-Meter (FOM) used to measure backfat and LM depth.
[5] Calculated by divided HCW by (percentage carcass yield divided by 100).
[6] Calculated by the difference between BW at the time of second Improvest® dose and harvest, divided by reported ADG, plus the age at second dose Improvest® dose.
[a,b,c] Within a row of a given variable, means without a common superscript differ ($P \leq 0.05$) as analyzed in respective cited studies.
[x,y] Within a row of a given variable, means without a common superscript differ ($P \leq 0.10$) as analyzed in respective cited studies.
X = Sex not included in the given study.
- = variable not observed in the study.

(2008) observed greater carcass fat-free lean in immunologically castrated pig compared with physically castrated pigs. Even within a study, use of different methods for determining carcass lean percentage leads to different estimated carcass lean conclusions. Boler et al. (2014) observed that immunologically castrated pigs have greater carcass fat-free lean percentage compared to physically castrated pigs when using the formula ([soft tissue weight (soft tissue weight × soft tissue % fat)]/chilled right side weight) × 100, but did not observe any differences when estimating carcass lean percentage using the Fat-O-Meter. Eleven different methods of estimating percentage carcass lean have been reported using different compositional variables, resulting in variable responses and conclusions about carcass composition among sexes (Table 1.4). As a result, it is difficult to confidently ascertain the carcass lean benefits of immunological castration. In general, immunological castration has a greater effect on decreasing backfat thickness than increasing longissimus muscle area or depth.

A slightly different approach for assessing carcass lean percentage is to determine the lean and cutting yield percentages (Table 1.4). This is based more on mass of primal cuts as a percentage of chilled side weight, rather than using linear measurements of backfat and longissimus muscle area or depth to calculate carcass lean percentage. In this context, immunologically castrated pigs had greater lean yield percentage (Boler et al., 2012; Pauly et al., 2009) and cutting yield percentage (Boler et al., 2012) compared to physically castrated pigs. However, in a subsequent study, lean and cutting yield percentages reported by Boler et al. (2014) were not different between immunologically and physically castrated pigs. This was consistent with the greater fat-free lean percentage of immunologically castrated pigs compared to physically castrated pigs, that

Table 1.4. Summary of carcass lean and lean and carcass yield percentages of intact male (IM), gilt (G), and physically castrated (PC) pigs compared with pigs immunologically castrated (IC) with Improvest®[1]

	Harvest, wk[2]			Carcass lean percentage				Lean yield percentage[3]				Cutting yield percentage[4]			
Study	BW at harvest, kg	Second Improvest® dose	Age	IM	G	PC	IC	IM	G	PC	IC	IM	G	PC	IC
Asmus et al. (2014)[5]	124.7[13]	5	25	X	X	54.3	55.5	X	X	-	-	X	X	-	-
Asmus et al. (2014)[5]	128.6[13]	7	28	X	X	52.2	52.3	X	X	-	-	X	X	-	-
Boler et al. (2014)[6]	135.7	5,6,7	25,26,27	64.5^{c}	60.6^{b}	58.1^{a}	61.1^{b}	64.7^{c}	62.8^{b}	61.5^{a}	62.5^{ab}	75.8^{c}	74.6^{b}	73.6^{a}	74.3^{ab}
Boler et al. (2014)[7]	135.7	5,6,7	25,26,27	51.8^{c}	49.9^{b}	48.3^{a}	48.2^{a}	-	-	-	-	-	-	-	-
Batorek et al. (2012)[8]	107.2	5	24	62.0^{b}	X	58.3^{a}	59.6^{a}	-	X	-	-	-	X	-	-
Boler et al. (2012)[7]	124.5	4	23	X	X	55.6^{y}	56.2^{x}	X	X	62.4^{a}	65.0^{b}	X	X	74.2^{a}	76.7^{b}
Boler et al. (2012)[7]	127.8	6	25	X	X	55.7^{y}	56.3^{x}	X	X	74.2^{a}	76.7b	X	X	74.2^{a}	76.3^{b}
Yuan et al. (2012)[9]	99.2	4	25	X	X	-	-	X	X	59.6	59.8	X	X	-	-
Pauly et al. (2009)[10]	107.0	5[14]	23[14]	-	X	-	-	57.5^{a}	X	54.5^{c}	56.3^{b}	-	X	-	-
Zamaratskaia et al. (2008)[11]	125.1	4	24	57.8^{a}	X	54.9^{b}	56.1^{b}	-	X	-	-	-	-	-	-
Zamaratskaia et al. (2008)[12]	125.1	4	24	60.2^{a}	X	56.5^{b}	58.5^{c}	-	X	-	-	-	-	-	-

[1] Studies that included the interaction between Improvest® and other nutrient partitioning agents (somatropin and ractopamine), did not provide sufficient information to determine the interval between the second Improvest® dose, or did not specify the use of the commercially available Improvest® were excluded from this summary.
[2] When age was given in d, age was converted to wk and rounded up to the whole wk.
[3] Lean cutting yield = [(trimmed ham + trimmed loin + butt shoulder + picnic)/ chilled side weight] × 100; except Pauly et al. (2009), see footnote 10
[4] Carcass cutting yield = [(trimmed ham + trimmed loin + butt shoulder + picnic + trimmed belly)/ chilled side weight] × 100.
[5] Percentage lean = 58.86 – (backfat × 0.61) + (loin depth × 0.12).
[6] Fat-free lean percentage = {[soft tissue weight (soft tissue weight × soft tissue % fat)]/chilled right side weight} × 100.
[7] Carcass lean percentage estimated using Fat-O-Meter.
[8] Lean meat content percentage = 60.81879 – 0.72992 × backfat (mm) + 0.12127 × loin depth (mm).
[9] Lean meat yield percentage = Lean meat weight/ (bone weight + lean meat weight + fat weight + skin weight) × 100.
[10] Lean meat percentage = sum of denuded shoulder, back and ham weights as percentage of cold carcass weight.
[11] Lean meat content percentage, commercial grading – Hennessy Grading Probe.
[12] Lean meat content percentage, estimated = (0.729 × % meat and bone in ham), lean and bone in ham used to estimate whole carcass.
[13] Calculated by divided HCW by (percentage carcass yield divided by 100).
[14] Calculated by the difference between BW at the time of second Improvest® dose and harvest, divided by reported ADG, plus the age at second dose Improvest® dose.
[a,b,c] Within a row of a given variable, means without a common superscript differ ($P \leq 0.05$) as analyzed in respective cited studies.

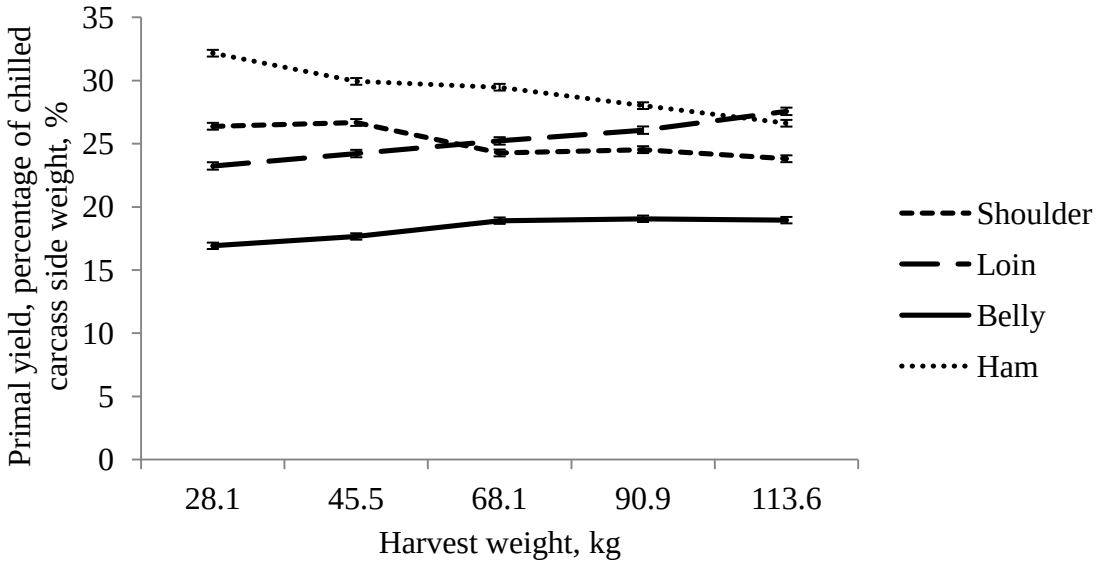
[x,y] Within a row of a given variable, means without a common superscript differ ($P \leq 0.10$) as analyzed in respective cited studies.
X = Sex not included in the given study.
- = variable not observed in the study.

was observed in the same study when using the Fat-O-Meter (Boler et al., 2014). Even with the variable responses in carcass lean percentage reported among studies, it is important to consider adipose and lean composition differences among carcass locations because primal cuts vary in economic value.

3. Carcass primal cut composition

The pork carcass can be segmented into various sections called primal cuts, which include the ham, loin, belly, butt shoulder, and picnic shoulder (IMPS, 2014). These primals can further be cut into subprimals and eventually sold as retail cuts. Each primal contributes different amounts to the overall carcass value, which is also known as the cutout value. Primals and subprimals have specifications that are used to standardize meat cutting consistency of muscles, fat, and bone (IMPS, 2014). These IMPS specifications influence the yield of each primal cut, as well as the mass and proportion of lean and fat of each primal and subprimal cut. Primal yield and the proportions of lean and fat change with increasing harvest weight. For example, the percentage of carcass weight contributed by the ham and shoulder decreases, and the percentage represented by the loin and belly increases as harvest weight increases (Figure 1.6; Apple et al., 2009b). Each unit of lean and fat in a primal cut has a disproportionate effect on the value of the primal. For example, the specification for IMPS #410 pork loin indicates that no more than 0.10 cm of backfat remains over the loin. The weight of 0.10 cm backfat that remains on the pork loin has a much greater value, than the remaining backfat over the loin that goes into trim. In contrast, carcass fat deposited in the belly primal is not discriminated against, and adds weight and thus, value to this primal cut. In regard to the resulting differences in lean and fat proportions to meet primal cut specifications, and the

Figure 1.6. Percentage primal yield of the shoulder, loin, ham, and belly of pigs from 28.1 to 113.6 kg



Adapted from (Apple et al., 2009b).

added-value of fat in the belly compared with other primal cuts, understanding lean and fat primal composition is necessary to maximize cutout value.

a. Ham, shoulder, and loin primal cuts

Studies have been conducted to determine primal weight differences and the proportion of lean and fat in pork carcasses from immunologically castrated pigs. Immunologically castrated pigs have a greater proportion of the carcass represented in the ham and whole shoulder (Pauly et al., 2009; Boler et al., 2012), butt shoulder and picnic shoulder (Boler et al., 2012) than physically castrated pigs. When the butt shoulder and picnic shoulder primal cuts were further fabricated to boneless primals, they continued to represent a greater portion of carcasses from immunologically castrated pigs compared to carcasses of physical castrates (Boler et al., 2012). Despite a lack of whole primal ham weight differences observed by Gispert et al. (2010) hams from immunologically castrated pigs had a greater proportion of lean and reduced proportion of fat compared to

physically castrated pigs. Thus, greater ham lean of immunologically castrated pigs and greater subcutaneous fat of physical castrates offset each other and resulted in similar whole ham weight.

Similar to the compositional makeup of the ham, Boler et al. (2012) and Gispert et al. (2010) observed that physically castrated pigs had a greater proportion of the carcass represented by the loin than immunologically castrated pigs. Upon further fabrication, the increased loin proportion in the carcass was due to greater proportion of fat in the physically castrated pigs compared to immunologically castrated pigs (Gispert et al., 2010, Boler et al., 2012). Producing pigs with more lean and less fat is beneficial in producing greater quantities of lean meat and thus primal cuts have greater value.

b. Belly primal cut

The belly represents a lower proportion of the carcass weight in gilts and physically castrated pigs than the ham, loin, and shoulder (Figure 1.6; Apple et al., 2009b). In all primal cuts, the percentage of dissected fat increases with increasing carcass weight from 17.8 to 80.6 kg (Apple et al., 2009b), but the belly primal cut has the greatest percentage of carcass fat (7.0%) compared to the shoulder, ham, and loin primal cuts (4.3, 4.1, and 6.3 percentage carcass fat, respectively; Apple et al., 2009b). Immunological castration decreases the proportion of the belly weight relative to carcass weight compared to physically castrated pigs (Pauly et al., 2009; Boler et al., 2012). The decrease in belly weight of immunologically castrated pigs was also accompanied by decreased belly width and thickness compared to physically castrated pigs (Boler et al., 2012). Increasing the time interval between the second Improvest® dose and harvest from 4 to 6 weeks (3.2 kg greater body weight), did not increase belly weight, width or

thickness. Emphasis today is placed on quality of pork fat quality which is heavily influenced by the amount and composition of dietary lipids (Wood et al., 2008). Primal cuts that contain high fat content (belly) are of particular concern regarding pork fat quality. Given the decreased belly thickness of immunologically castrated pigs and the inability to increase belly thickness by increasing the interval between the second Improvest® dose and harvest, feeding diets high in PUFA content could be detrimental to belly quality of immunologically castrated pigs.

F. Considerations for carcass lean and fat quality

1. Animal behavior and premortem handling

Intact male pigs have more skin abrasions and blemishes than gilts and physical castrates (Babol and Squires, 1995). These defects can be superficial, or result in muscle bruising which subsequently results in carcass trimming and loss in value to pork packers and producers (Babol and Squires, 1995). Results from a recent evaluation of transportation welfare of market pigs showed that the percentage of pigs dead on arrival and non-ambulatory pigs decreased from 1.17% for physically castrated pigs to 0% for immunologically castrated pigs upon arrival at a commercial abattoir (Guay et al., 2013). Transport mortality can occur for a myriad of reasons and results in additional financial losses to producers and packers (Johnson et al., 2013). Evaluation of possible physiological changes of immunologically castrated pigs during transport has not been conducted, but physically castrated and immunologically castrated pigs should behave similarly after the second dose of Improvest®. Therefore, it is unclear why immunologically castrated pigs seem to cope with transportation stress more successfully than physically castrated pigs. Perhaps the increased aggressive behavior of

immunologically castrated pigs before the second Improvest® dose allows pigs to adapt to periods of stress more effectively after the second Improvest® dose. Many antemortem and postmortem factors, including transportation, influence the postmortem pH decline of muscle, which determines postmortem muscle metabolism and influences lean quality (Berg and Eilert, 1995). Defining pork quality can be a contentious topic in a pork production "culture" where quantity has taken precedence over quality. The emphasis on increased pork carcass lean in the U.S. has been dictated by consumer demand for leaner meat and has resulted in carcass price premiums paid by pork processors to promote greater carcass lean and less fat. Ultimately, "quality" is a relative assessment and has been defined as the relationship between desired and perceived properties of a given product, measured by the degree of satisfaction through the point of consumption (Hugo and Roodt, 2007).

Meat quality encompasses sensory characteristics of lean and fat that affect consumer acceptance, such as boar taint aroma, as well as other off-flavors and aromas related to lipid peroxidation (Babol and Squires, 1995). In general, pork lean quality is classified as either acceptable or unacceptable based on the presence of pale, soft, and exudative (**PSE**), or dark, firm, and dry lean (Aberle et al., 2001). The PSE quality defect has been a problem in the U.S. pork industry for many years (Klinkner et al., 2013). Typically, measures of lean quality have been focused on water holding capacity, color, firmness, and texture which consumers evaluate through visual, tactile, and gustatory senses at the point of purchase, and these measures contribute to sensory attributes of tenderness, juiciness, and flavor upon consumption (Aberle et al., 2001). These quality characteristics in fresh pork products determine the likelihood of a customer returning to

purchase pork products after their previous experience. Lean quality of pork is most commonly determined using the pork loin due to its ease of accessibility for evaluation, economic value, glycolytic activity, and sensitivity to changes in postmortem metabolism during the conversion of muscle to meat.

2. Lean composition and quality

Lean that is classified as pale, soft, and exudative is closely linked to a short-term stress prior to harvest, and dark, firm, and dry is closely associated with long-term stress and the corresponding changes to glucose metabolism and pH decline (Lawrence et al., 2012). A literature review conducted by Babol and Squires (1995) indicated that in most cases, there is no difference in the occurrence of pale, soft, and exudative between intact male, physically castrated, and gilt pigs. However, intact male pigs can be more susceptible to developing dark, firm, and dry lean than physical castrates and gilts (Babol and Squires, 1995). Intact male pigs are more aggressive throughout the finishing period (Guay et al., 2013), which constitutes a long-term stress and can result in depletion of muscle glycogen before harvest, thus limiting postmortem pH decline (Aberle et al., 2001). Immunological castration has very little effect on postmortem metabolism related to pH decline, as indicated by similar 45 min postmortem pH and ultimate pH of the longissimus muscle (Gispert et al., 2010; Pauly et al., 2009; Zamaratskaia et al., 2008a; Boler et al., 2012; Boler et al., 2014), the semimembranosus muscle (Gispert et al., 2010), and ultimate pH of the biceps femoris (Zamaratskaia et al., 2008a) among immunological castrates, intact males, gilts, and physical castrates.

Lean color of meat can be assessed subjectively and objectively. Objective color measurement involves the use of Hunter or Minolta colorimeters to determine L* (0 =

black and 100 = white), a* (a more negative value represents more green lean and a more positive value represents more red), and b* (a more negative value represents more blue and a more positive value represents more yellow). Objective color (L*, a*, and b*) of the longissimus muscle using a Hunter (Pauly et al., 2009) or Minolta (Boler et al., 2012; Boler et al., 2014) colorimeter was not different between intact males, immunological castrates, or physical castrates (Boler et al., 2012; Pauly et al., 2009) and gilts (Boler et al., 2014). In contrast, Gispert et al. (2010) reported that the semimembranosus muscle was slightly lighter in color (higher L*) from immunologically castrated pigs compared to intact male pigs, but similar to physically castrated and gilt pigs (Gispert et al., 2010). In any case, differences in subjective color evaluation were not detected among sexes (Gispert et al., 2010; Boler et al., 2012; Boler et al., 2014).

Increasing the time interval between the second Improvest® dose and harvest from 4 to 6 weeks did not alter longissimus muscle L* values of immunologically and physically castrated pigs, but increasing the time interval increased longissimus muscle a* and b* (Boler et al., 2012). The increase in a* also occurred in physically castrated pigs, and since myoglobin content of muscle increases with advancing chronological age (Aberle et al., 2001), this explains the increase in a* value that occurs in both immunologically and physically castrated pigs.

Water holding capacity of lean is a function of the moisture content, and the ability of muscle to retain moisture declines as pH declines and nears the isoelectric point of meat (Aberle et al., 2001). Since intact male pigs have less lipid and greater moisture content compared to physically castrated pigs (Nold et al., 1997), it is not surprising that the longissimus muscle of immunologically castrated pigs has greater moisture and

reduced lipid content compared to physically castrated pigs (Boler et al., 2012; Boler et al., 2014). While the moisture content of the longissimus muscle of immunologically castrated pigs decreased, and the lipid content increased, by increasing the time period between harvest and the second Improvest® dose from 4 to 6 weeks, pigs harvested at 4 or 6 weeks after the second Improvest® dose had greater moisture and less lipid compared to physically castrated pigs (Boler et al., 2012). Boler et al. (2012) observed no differences in drip loss percentage of the longissimus muscle between immunologically and physically castrated pigs. Since water adds to the primal weight, primal value should be increased in immunologically castrated pigs that have greater moisture content and no change in water holding capacity.

Other sensory attributes can be objectively measured. Shear force can be used to measure perceived tenderness (Lawrence et al., 2012). Shear force tends to be greater in immunologically castrated pigs compared to physically castrated pigs of the same age (Boler et al., 2012). However, others have shown that shear force is reduced in immunologically castrated pigs compared to physically castrated and intact male pigs (Pauly et al., 2009). This difference between these 2 studies is likely due to muscle composition in relation to the reference comparisons. Collagen content reduces meat tenderness (Aberle et al., 2001). Intact male pigs have more collagen than physically castrated and gilt pigs (Nold et al., 1999) due to androgens stimulating collagen synthesis (Claus et al., 2007). However, following the second Improvest® dose, plasma hydroxyproline decreases, implying collagen breakdown (Claus et al., 2007). The only way to overcome decreased tenderness due to collagen deposition, is through increased lean deposition to cause dilution (Aberle et al., 2001). Since Boler et al. (2012) marketed

pigs at a heavier body weight, older-age, and increased interval between the second Improvest® dose and harvest compared to Pauly et al. (2009), the dilution affect would have likely been greater, resulting in similar shear force in immunological castrates compared to physically castrated pigs. Decreased tenderness could be one reason to market pigs with longer intervals between the second Improvest® and harvest, especially in heavy weight pigs. Lean color, pH, and measures of water holding capacity are used to indirectly measure postmortem metabolism and objectively relate to subjective visual appearance and palatability characteristics.

Objective quality characteristics have been consistent with lower subjective marbling scores and slightly lower firmness scores of loins from immunologically castrated pigs compared to loins from physically castrated pigs (Boler et al., 2012). However, other studies have reported no difference in subjective quality characteristics of the longissimus muscle among immunologically castrates, intact males, physical castrates, and gilts (Boler et al., 2014). The inconsistent subjective quality differences of the longissmus muscle between the studies by Boler et al. (2012) and Boler et al. (2014) are possibly due to greater adiposity (backfat thickness) of pigs in Boler et al. (2014) study as a result of longer time intervals between the second Improvest® dose and heavier body weight at harvest. Since immunologically castrated pigs have less carcass fat than physically castrated pigs, the compositional changes of carcass fat are more likely to be altered.

3. Pork fat quality

a. Changing consumer preferences for fat

Consumer preferences for fat have changed over time. In the early 1900's pork fat was valued for cooking, but more recently, pork fat is perceived as unhealthy (NPB, 2013; De Smet et al., 2004). To meet consumer preference for meat with less fat, swine genotypes have been selected for increased lean and reduced fat (De Smet et al., 2004). Today, pork contains 16% less fat than 20 years ago, and 75% less fat than 60 years ago (NPB, 2013). While pork packers, processors, and producers have responded to the consumer desire for leaner pork, it has led to complaints of increased lean and fat separation, decreased production yields, and increased "greasing out" and smearing of fat, all indicators of poor fat quality (Apple, 2010).

b. Assessment of pork fat quality

"Quality" is a subjective assessment that depends upon person-specific desired characteristics (Hugo and Roodt, 2007). Wood et al. (1984) provided a generalized description of poor fat quality as "soft, oily, wet, grey and floppy". Quantification methods of fat quality have varied from subjective to objective methods, and the characteristics assessed have ranged from textural to chemical (Table 1.5). Early studies that evaluated soft pork fat reported saponification value, specific gravity, iodine value, refractive index, melting point, titer, and fatty acid composition (Ellis and Isbell, 1926). Additional methods have been developed to determine fat quality, and in many cases, relate fat quality to belly firmness (Table 1.5). The most widely used assessment method today is fatty acid composition and calculated iodine value (Table 1.5). Fatty acids can differ in carbon chain length and degree of unsaturation. Increasing the number of double

Table 1.5. Methods of determining pork fat quality and fresh belly quality

	Rating scale		Chemical composition				Fat color			Belly					
Study	Subjective	Objective	Proximate	Fatty acid	Calculated IV[1]	Melting point	Subjective – JCS[2]	Objective – colorimeter	Thickness	Flop angle	Flex angle	Flop distance	Compression	Puncture	Slicing yield
Barton-Gade, 1987			X	X	X										
Miller et al., 1990	X			X											
Sather et al., 1995		X													
Averette Gatlin et al., 2002				X	X	X						X	X		
Apple et al., 2007				X	X			X	X			X	X		
Correa et al., 2008	X			X	X										
Leick et al., 2010				X	X			X	X			X			
Meadus et al., 2010		X													
Xu et al., 2010b				X	X		X	X	X	X					
Xu et al., 2010a				X	X		X		X	X					
Dahlen et al., 2011				X	X	X			X	X					
Trusell et al., 2011			X	X	X				X	X		X	X	X	
Boler et al., 2012				X	X				X			X			
McClelland et al., 2012				X	X				X		X				X
Hilbrands et al., 2013				X	X				X						
Lee et al., 2013								X				X			
Tavárez et al., 2014a				X	X				X			X			X
Tavárez et al., 2014b			X	X	X										
Kyle et al., 2014				X	X				X			X			X

[1] IV = Iodine value

[2] JCS = Japanese color score

bonds in the carbon chain increases the degree of unsaturation, and determines the physical characteristics of lipids (Azain, 2001). For example, increasing the chain length and degree of unsaturation of fatty acids decreases the melting point and causes lipids to be in liquid form at room temperature (Azain, 2001).

Iodine value was originally developed as a wet chemistry procedure in the late 1800's (AOCS, 2009) to measure the degree of fatty acid unsaturation defined as the number of grams of iodine absorbed by 100 g of lipid (Shahidi and Wanasundara, 2008a). This method has limitations including time-consuming methodology and use of harmful reagents (Kyriakidis and Katsiloulis, 2000). With advances in gas chromatography, the American Oil Chemists Society developed a method of calculating iodine value to overcome the limitations of the wet chemistry iodine value procedure. In the AOCS (1998) method, iodine value is calculated from the fatty acid composition of lipids and uses different coefficient weights on specific fatty acids (Table 1.6; Kyriakidis and Katsiloulis, 2000). The AOCS (1998) iodine value equation has been widely applied to pork fat quality determination. Meadus et al. (2010) first reported the use of a different equation that included additional fatty acids than included in the AOCS (1998) equation. The additional fatty acids have longer carbon chains (C20 and C22 fatty acids) with more double bonds (greater unsaturation; Table 1.6). Development of the Meadus iodine value is not well documented, and is less commonly used than the AOCS (1998) iodine value equation. However, the C20 and C22 fatty acids are important to consider because more unsaturated, longer carbon chain fatty acids have more interrupted methylene groups (double bonds). As the number of interrupted methylene groups increases, less energy is required for hydrogen abstraction by free radicals (Wood et al., 2008). Thus, more

Table 1.6. Comparison of fatty acids and coefficients of fatty acids included in two different iodine value (**IV**) equations

Fatty acid	IV-AOCS[1]	IV-Meadus[2]
C16:1c	0.950	0.950
C18:1n-9	0.860	0.860
C18:2n-6	1.732	1.732
C18:3n-3	2.616	2.616
C20:1n-9	0.785	0.795
C20:2	--	1.570
C20:3n-3	--	2.380
C20:4n-6	--	3.190
C20:5n-3	--	4.010
C22:1n-9	0.723	--
C22:4n-6	--	2.930
C22:5n-3	--	3.680
C22:6n-3	--	2.930

[1] (AOCS, 1998)
[2] (Meadus et al., 2010)

unsaturated fatty acids are more susceptible to peroxidation. Oleic, linoleic, and linolenic acids contain 1, 2, and 3 double bonds, respectively, and as a result, peroxidation rate of these fatty acids occurs at a ratio of 1:12:25, respectively (Kim and Min, 2008). In addition, location of the interrupted methylene group influences the rate of peroxidation, where n-3 fatty acids oxidize at a faster rate than n-6 fatty acids (Erickson, 2008). Since the Meadus equation includes fatty acids that are structurally more susceptible to peroxidation, use of this equation may distinguish the degree of pork quality deterioration due to peroxidation better than the AOCS (1998) equation.

Currently, packers do not have a rapid, non-invasive, and inexpensive method to measure fat quality in pork carcasses. Most recent advances for determining pork fat quality in pork packing plants have been focused on the use of near infrared spectroscopy (Sørensen et al., 2012). In the meantime, some U.S. packers have established arbitrary acceptability thresholds and randomly survey suppliers. Suggested acceptable pork fat

quality thresholds have ranged from an IV of 70 in backfat (Barton-Gade, 1987) to an IV of 75 in backfat (Boyd et al., 1997), and most recently 73 in jowl fat (Benz et al., 2011b). The rationale for using these suggested standards relative to potential economic losses due to reduced pork fat quality are unclear.

For the last few decades, packers have communicated their desired procurement specifications by using carcass lean-based premium and discount incentives, which include some measure of carcass lean and fat (e.g. loin depth and/or backfat thickness), and hot carcass weight. Development of fat quality assessment methods that are in real-time, rapid, and non-destructive to major primal cuts would allow the use of fat quality as an additional procurement specification. Even with adoption of near infrared spectroscopy in pork packing plants, there is no standard carcass sampling location. Sampling site is important because fatty acid composition varies among carcass fat depots.

c. Fatty acid compositional differences among depots

Multiple pork fat depots are routinely assessed to determine fatty acid composition and calculate iodine value in an attempt to relate fat depot composition to belly quality. Determining fatty acid composition of belly fat is laborious, expensive, and requires excising a portion of the belly which would cause destruction and devaluation of the belly primal. The advantage of using backfat and jowl fat is they are easily accessible and do not result in devaluation of primal cuts. However, the lipogenic activity is lower in jowl fat than backfat and belly fat leading to different contributions of dietary fatty acids to the overall fatty acid composition of the fat depot (Mourot et al., 1995). When

lipogenic activity is low, dietary fatty acid composition has a greater effect on fatty acid composition of pork fat. Consequently, the fatty acid composition of pork fat varies across tissues, fat depots, depot location, and within primals.

1. Classes and functions of lipids

All lipids are insoluble in water but are classified based on chemical structure, physical properties, and biological function (O'Keefe, 2008). Fats and oils are storage lipids in the form of fatty acid(s) esterified to a 3-carbon glycerol backbone. Phospholipids and sterols are structural forms of lipids, and phosphatidylinositol, eicosanoids, and steroid hormones (including testosterone, estradiol, and androstenone) serve as intracellular, paracrine, and endocrine signaling lipids (O'Keefe, 2008). In the pork carcass, the predominant lipid classes in fat are triglycerides while phospholipids are the predominant lipid class in muscle (Wood et al., 2008). These lipid classes have the greatest influence on fatty acid composition among tissues and fat depots.

2. Fatty acid content of longissimus muscle vs. subcutaneous fat

The total fatty acid content of pork carcass subcutaneous fat is much greater than the total fatty acid content of the longissimus muscle (Wood et al., 2008). Both of these tissues (subcutaneous fat and longissimus muscle) are comprised predominately of oleic (35.8 and 32.8 g/100g of fatty acids, respectively) and palmitic acids (23.9 and 23.2 g/100g of fatty acids, respectively; Wood et al., 2008). However, subcutaneous fat contains much lower concentrations of arachidonic and eicosapentaenoic acids than the longissimus muscle (Wood et al., 2008). The fatty acid composition differences between subcutaneous fat and longissimus muscle can be attributed to a greater proportion of phospholipids compared to triglycerides in muscle, and a greater proportion of

triglycerides compared to phospholipids in fat (Wood et al., 2008). The phospholipid fraction has a lower concentration of oleic acid, but a greater concentration of linoleic, arachidonic, and eicosapentaenoic acids compared to the triglyceride fraction (Wood et al., 2008; Warnants et al., 1999; Bee et al., 2002).

3. Fatty acid composition differences among fat depots

Priority of fat deposition is given to internal fat depots, followed by subcutaneous, intermuscular, and intramuscular fat depots (Aberle et al., 2001). An example of internal fat deposition is the omental fat depot, which has a more saturated fatty acid composition compared to subcutaneous backfat due to lower oleic acid and greater palmitic acid (Bee et al., 2002). Another internal fat depot, leaf fat (kidney fat), also has greater total saturated fatty acid content compared to intramuscular and backfat depots (Koch et al., 1968). Internal fat depots, such as leaf fat, have a greater lipogenic rate (Mourot et al., 1995) resulting in more saturated fatty acid content compared to other fat depots. Subcutaneous and intramuscular fat depots have a similar fatty acid profiles (Wiegand et al., 2011). Intramuscular fat has a lower lipid content, but the lipid content is highly unsaturated due to the higher proportion phospholipids compared with subcutaneous fat. In contrast, subcutaneous fat has a greater proportion of triglycerides which are less unsaturated, thus resulting in similar fatty acid composition between intramuscular fat and subcutaneous fat (Wood et al., 2008). Subcutaneous fat of the belly has been shown to have a higher iodine value, indicating greater amounts of unsaturated fatty acids compared to intermuscular fat (Wiegand et al., 2011). More importantly, when dietary treatments are imposed, changes in fatty acid composition are not consistent across adipose tissue depots (Apple et al., 2011; Wiegand et al., 2011).

4. Fatty acid composition differences among subcutaneous depots

In general, pork fat becomes more saturated as pigs become fatter (Apple et al., 2009b) by diluting the unsaturated phospholipid fraction when triacylgerides are deposited in adipocytes (Wood et al., 2008). Subcutaneous fat depots do not respond consistently to changes in dietary fatty acid composition. Duttlinger et al. (2012) observed that feeding 20% dried distillers grains with solubles (**DDGS**) resulted in an increase in iodine value of 4.5, 6.3, and 6.9 g/100g of jowl fat, belly fat, and backfat, respectively. This is slightly greater than the tissue specific incremental increases reported by Benz et al. (2010) where jowl fat, belly fat, and backfat iodine value increased 1.6, 2.2, and 2.3 g/100g for every 10% increase in DDGS inclusion. The unequal response to dietary changes in fatty acid composition is mostly attributed to changes in differences in *de novo* lipogenesis rate across fat depots. The rate of *de novo* lipogenesis is lower in the forequarter region compared to the belly, loin, and ham (Mourot et al., 1995). A lower rate of *de novo* lipogenesis means that the dietary fat composition will have a greater influence on fatty acid composition of carcass fat. Jowl fat is typically more unsaturated compared to belly, backfat (Asmus et al., 2014b, Browne et al., 2013), and clear plate fat (Asmus et al., 2014b). This suggests that the use one fat depot to estimate the fatty acid composition of another depot location lacks merit and could result in inaccurate quality classifications.

5. Fatty acid concentration differences within fat depot location

Fatty acid composition differs within primal and depot location such as the belly and layers of backfat (Trusell et al., 2011; McClelland et al., 2012). When the belly was divided into 15 different sections to determined fatty acid composition differences, the

degree of fatty acid unsaturation declines (iodine value becomes lower) across a gradient from the dorsal anterior region toward the ventral posterior region (Trusell et al., 2011). Backfat of market weight pork carcasses is composed of multiple layers. Some researchers have observed that the inner backfat layer contains 0.5 to 1.5% more unsaturated fatty acids than the outer backfat layer (McClelland et al., 2012), while others have observed 1.0 to 1.5% more unsaturated fatty acids in the outer backfat layer compared to the inner backfat layer (Bee et al., 2002). These conflicting results are possibly due to the inability to precisely separate backfat layers. In general, sampling location within fat depots is an important factor that must be considered when determining pork fat quality. This appears to be especially important when using diet feeding strategies that alter fatty acid composition of pork fat, since fat depots respond differently to the amount and composition of fatty acids in diets.

d. Carcass fat characteristics among sexes

Backfat from intact male pigs has less lipid and greater water content compared to physical castrates and gilts (Wood et al., 1989; Barton-Gade, 1987). Furthermore, backfat from intact male pigs has a lower percentage of oleic acid, and greater percentages of linoleic and linolenic acids, compared to physical castrates and gilts (Kyle et al., 2014; Wood et al., 1989; Barton-Gade, 1987). Subjective fat firmness measures have also been used to show that backfat from intact male pigs is softer than backfat from physically castrated pigs (Sather et al., 1995). It is expected that immunologically castrated pigs have a similar fatty acid composition and fat quality to intact male pigs, and this will be discussed later in this review.

e. Consequences of soft pork fat

Pork fat high in unsaturated fatty acids results in reduced firmness and causes poor handling characteristics, reduced slicing yield (Morgan et al., 1994), and decreased shelf-life due to lipid peroxidation (Wood et al., 2008). Although the majority of attention has been focused on dietary effects on pork fat quality (which will be discussed later), the greater concentration of unsaturated fatty acids in adipose tissue from intact male pigs could predispose immunologically castrated pigs to have greater reductions in pork fat firmness when fed diets containing large quantities of polyunsaturated fatty acids compared to physical castrates. These effects may be of the greatest concern in the belly primal, which contains the greatest proportion of adipose relative to lean among all primal cuts in the carcass (Figure 1.6; Apple et al., 2009b). The increased demand for bacon in the U.S. has dramatically increased the value of bellies, and as a result, is a major concern for pork packers and processors. Moreover, use of feed ingredients that have a high concentration of polyunsaturated fatty acids (e.g. corn dried distillers grains with solubles), have perpetuated pork fat quality challenges of lean pigs.

VI. Physiology of adipose accretion

Lipid in adipose tissue is a storage form of energy that accumulates when positive energy balance is achieved and energy consumption is greater than that required for maintenance and growth (Kersten, 2001; van Milgen and Noblet, 2003). Thus, the rate of adipose tissue accretion is the result of the balance between lipogenesis and lipolysis (Kersten, 2001). As a pig becomes fatter, and more lipid filling occurs in adipocytes, the percentage of lipid increases, while the percentage of water decreases in adipose tissue depots (Wood et al., 1989). Subcutaneous backfat measuring 8 mm contained 69.2% lipid

and 22.4% water, while pork carcasses with 12 and 16 mm of backfat had 77.0% and 81.6% lipid, respectively, and 17.1% and 14.1% water, respectively (Wood et al., 1989).

In pigs, lipid accretion in adipose tissue can occur from either digestion and absorption of dietary fatty acids, or by using glucose as a carbon source to synthesize fatty acids through *de novo* lipogenesis (Dodson et al., 2010). The balance between the rate of deposition from dietary fatty acids and *de novo* lipogenesis is regulated by enzymatic activity and is dependent on body weight, fat depot, sex, and diet composition (Hugo and Roodt, 2007).

A. Sources of fatty acids in pork adipose tissue

1. De novo lipogenesis

Lipogenic precursors and the site of lipogenesis differ among species (Dodson et al., 2010). In humans and rodents, lipogenesis occurs predominantly in the liver, while in pigs, lipogenesis occurs exclusively in adipose tissue (Dodson et al., 2010). Lipogenic enzymes regulate *de novo* lipogenesis (White et al., 2013). Acetyl CoA is produced during glycolysis from metabolism of carbohydrates, and is the initial substrate necessary for the lipogenic pathway to commence (White et al., 2013). Acetyl CoA carboxylase, is the rate-limiting lipogenic enzyme that converts acetyl CoA to malonyl CoA, and fatty acid synthase converts malonyl CoA to palmitate, which is a saturated fatty acid (White et al., 2013). Stearoyl-CoA desaturase, also called Δ9 desaturase, controls elongation and desaturation of monounsaturated fatty acids (**MUFA**) from acetyl-CoA (White et al., 2013). Elongation and desaturation of polyunsaturated fatty acids (**PUFA**) is catalyzed by Δ6 desaturase, elongase, and Δ5 desaturase from linoleic and linolenic acids results in the synthesis of arachidonic, eicosapentaenoic, and docosahexaenoic acids (White et al.,

2013; Farnworth and Kramer, 1987) which are known for their human health benefits (Swanson et al., 2012).

Lipogenic enzyme expression has been compared among intact males and immunologically and physically castrated pigs (Mackay et al., 2013). Subcutaneous adipose tissue of immunologically castrated pigs had similar steroyl CoA desaturatase and Δ6-desaturase expression compared to intact male pigs, but Δ6-desaturase was greater in immunologically castrated pigs compared to physical castrates (Mackay et al., 2013). Conversely, fatty acid synthase expression was similar between immunologically and physically castrated pigs, but both had greater fatty acid synthase expression compared with intact males (Mackay et al., 2013). Others have observed increased fatty acid synthase and acetyl CoA carboxylase in subcutaneous and perirenal fat depots of physical castrates compared to intact male pigs at 147 and 210 days of age (Yao et al., 2011). The reduced lipogenic rate of intact male pigs compared to physical castrates is due to increased circulating testosterone of intact males compared to physical castrates (Yao et al., 2011). The decreased lipogenic enzyme rate of intact males suggests lower *de novo* lipogenesis and a greater proportion of fatty acid deposition derived from the diet and smaller proportion of fatty acids derived from *de novo* lipogenesis.

Mackay et al. (2013) did not evaluate the expression of acetyl CoA carboxylase, which is the rate limiting enzyme for *de novo* lipogenesis. The pigs used in their study were harvested over a 3 to 6 week period after the second Improvest® dose, but this was not considered in the data analysis. Hot carcass weight of these pigs was about 100 kg, which is typical in the European Union, but much less than pigs harvested in the U.S. This is important to note because lipogenic enzyme expression changes at varying rates

among adipose tissue depots as body weight increases (Mourot et al., 1995). Acetyl CoA carboxylase activity in the belly does not change from 20 to 120 kg body weight, while acetyl CoA carboxylase activity of backfat, neck, ham, throat, and leaf fat does not change after 80 kg body weight (Mourot et al., 1995). Fat accretion in adipose tissue continues with advancing age despite the general decrease lipogenic enzyme activity (Farnworth and Kramer, 1987). Thus, fatty acids must come from another source. It is unknown how lipogenic enzyme expression changes as the time period between the second Improvest® dose and harvest is increased, but studies have characterized the fatty acid profile of adipose tissues during this the time period (Table 1.7; Asmus et al., 2014b; Kyle et al., 2014; Tavárez et al., 2014b; Boler et al., 2012). Saturated fatty acid content of belly fat was increased (as indicated by lower iodine value) when the interval between second Improvest® dose and harvest was increased from to 2 to 4 weeks, but no further changes were observed when increasing the interval to 6 or 8 weeks (Tavárez et al., 2014b). However, an increase in saturated fatty acids of belly fat was observed when the interval between second Improvest® dose and harvest was increased from 4 to 6 weeks (Boler et al., 2012). This increase in saturated fatty acid content is likely a function of greater fat accretion and lipid filling that occurs by extending the interval between the second Improvest® dose and harvest. Thus, the increasing the interval between the second Improvest® dose and harvest would improve pork fat quality of immunologically castrated pigs. However, it is common to include dietary lipids high in PUFA content, in swine diets, which can also decrease pork fat quality (Apple et al., 2009a).

Table 1.7. Summary of fatty acid composition and iodine value[1] of belly adipose tissue from intact male (IM), gilt (G), physically castrated (PC), and immunologically castrated (IC) pigs

		Boler et al. (2012)		Asmus et al. (2014)[2]		Travarez et al. (2014)				Kyle et al. (2014)
General diet formulation		Corn-soybean meal with 20% DDGS[3] until 21 WOA[4] then 10% DDGS		Corn-soybean meal with either 0% DDGS, 30% DDGS, or 30% DDGS with a 5 or 7 wk withdrawal		Corn-soybean meal				No diet information
BW at harvest, kg		124.5	127.8	124.7[5]	128.6[5]	~120	~128	~140	~144	135.7
Interval between second Improvest® dose and harvest, wk		4	6	5	7	2	4	6	8	5,6,7
Age at harvest, wk		23	25	25	28	22	24	26	28	25, 26, 27
C16:0	IM	X	X	X	X	X	X	X	X	20.08[abc]
	G	X	X	X	X	X	X	X	X	19.55[a]
	PC	22.68	22.32	23.4	23.33	27.35	26.55	25.85	26.87	20.60[c]
	IC	22.54	22.24	23.1	23.63	27.76	26.02	25.48	22.52	20.54[bc]
C18:0	IM	X	X	X	X	X	X	X	X	10.64[a]
	G	X	X	X	X	X	X	X	X	11.86[b]
	PC	7.70[6]	8.05	11.56	11.59	13.90	14.02	11.41	11.66	11.66[b]
	IC	7.62[6]	8.75	12.20	12.25	13.08	11.55	14.62	12.48	11.26[ab]
C18:1	IM	X	X	X	X	X	X	X	X	42.51[a]
	G	X	X	X	X	X	X	X	X	44.77[b]
	PC	40.52[b6]	41.80[b]	37.56	38.28	37.42	39.14	41.45	38.55	44.89[b]
	IC	39.02[a6]	40.15[a]	36.59	37.66	37.79	39.05	40.09	41.21	44.48[b]

C18:2	IM	X	X	X	X	X	X	X	X	16.06[c]
	G	X	X	X	X	X	X	X	X	14.56[b]
	PC	19.36[a6]	18.20[a]	15.21	14.73	13.40[a6]	12.76[a]	12.12[a]	12.92[a]	13.41[a]
	IC	21.44[b6]	19.47[b]	16.23	14.60	16.29[b6]	13.85[b]	13.35[b]	14.31[b]	13.90[ab]
C18:3	IM	X	X	X	X	X	X	X	X	0.85[b]
	G	X	X	X	X	X	X	X	X	0.74[a]
	PC	0.89	0.90	0.58	0.55	0.63[a6]	0.58[a]	0.55[a]	0.57[a]	0.69[a]
	IC	1.05	0.85	0.62	0.54	0.72[b6]	0.64[b]	0.60[b]	0.66[b]	0.71[a]
C20:4	IM	X	X	X	X	X	X	X	X	0.09
	G	X	X	X	X	X	X	X	X	0.05
	PC	0.43[6]	0.38	0.25	0.25	0.36	0.35	0.33	0.36	0.02
	IC	0.45[6]	0.41	0.30	0.25	0.43	0.33	0.32	0.40	0.02
Iodine value[1]	IM	X	X	X	X	X	X	X	X	70.6[b]
	G	X	X	X	X	X	X	X	X	69.5[a]
	PC	77.7[ax]	77.2[ay]	66.8	66.5	~62[a]	~61	~61	~61	67.6[a]
	IC	79.9[bx]	78.0[by]	67.5	65.5	~67[b]	~64	~64	~61	68.3[a]

[1] Iodine value = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + ([C22:1] × 0.723) (AOCS, 1998).
[2] Means of 3 experimental dietary treatments pooled.
[3] DDGS = corn Dried distillers grains with solubles.
[4] WOA = wk of age.
[5] Calculated by divided HCW by (percentage carcass yield divided by 100).
[6] Significant effect ($P \leq 0.05$) by extending the time interval between the second Improvest® dose and harvest.
[a,b,c] Within a column of a given variable, means without a common superscript differ ($P \leq 0.05$) as analyzed in respective cited studies.
[x,y] Within a row of a given variable, means without a common superscript differ ($P \leq 0.10$) as analyzed in respective cited studies.
X = Sex not included in the given study.
~ Means are estimated from graphical display.

2. Dietary lipids

a. Benefits of dietary lipids

Dietary lipids (animal fats, vegetable oils, and blends) have been added to swine diets for many years as a rich source of energy to increase the energy density of the diet (Patience, 2012). Lipid addition to swine diets is particularly important during the hot summer months when feed intake decreases (Patience, 2012). This seasonal reduction in feed intake and growth rate routinely results in decreased hot carcass weight from June-July in the U.S. (Meyer and Steiner, 2014c). Dietary lipids have a lower heat increment, which reduces heat loss due to digestion and results in greater metabolizable energy utilization efficiency (percentage of metabolizable energy that is net energy), compared to starch, dietary fiber, and crude protein (90%, vs. 80%, vs. 60%, vs. 60%, respectively; Patience, 2012).

The addition of fat to diets increases the production rate of pelleting, but decreases pellet durability especially when adding more than 2% to the diet (Fairfield, 2005). Adding lipids to swine diets reduces dust (NRC, 2012), which increases equipment longevity and improves air quality in confinement swine facilities (Rosentrater, 2003). Lipids also provide essential fatty acids (i.e. linoleic, linolenic, and arachidonic acids) which cannot be synthesized, or synthesized in large enough quantities to meet requirements of the pig, and are critical for many biological functions (NRC, 2012).

b. Sources of dietary lipids

Dietary fats originate from animal and plant sources (NRC, 2012). These broad classifications have different energy values and fatty acid composition (Table 1.8; NRC,

2012). Animal-derived fat sources have a more saturated fatty acid profile and thus a higher melting point, lower titer (temperature where fat is completely solid), and lower iodine value (Azain, 2001; NRC, 2012). Choice white grease (rendered pork fat) is a common saturated fat source used in the U.S. swine industry (Azain, 2001). Plant-based fat sources have a greater proportion of unsaturated fatty acids, and thus lower melting point, higher titer, and higher iodine value (generally > 100; NRC, 2012) than animal fats. The two most common vegetable oils used in swine diets in the U.S. are soybean oil and corn oil. Corn contains 3.48% ether extract (NRC, 2012), which is comprised of 53.5% linoleic and 1.16% linolenic (Table 1.8; NRC, 2012). Corn oil does not contain any arachidonic acid (C20:4n-6), but it can be synthesized from linoleic and linolenic acid by the pig (White et al., 2013). In contrast to corn oil, choice white grease and beef tallow contain more saturated fatty acids (palmitic, steric, and oleic) but less metabolizable energy (Table 1.8; NRC, 2012). Typically, selection of lipid sources is based on handling characteristics and cost per unit of energy (NRC, 2012).

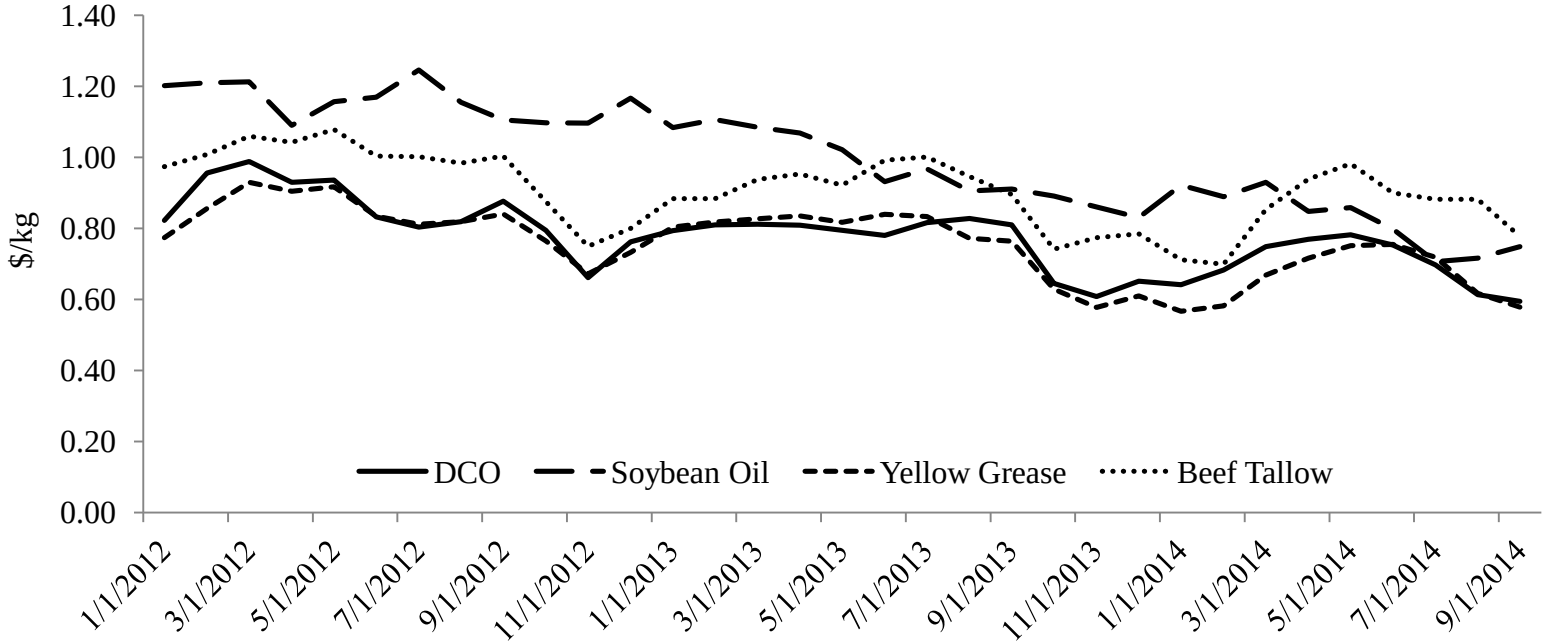
Anecdotally, several key factors have contributed to growing use of corn oil in swine diets including: minimizing the potential risk of disease transmission (Shoen, 2014), the improved digestibility of extracted corn oil compared to 'intact' corn oil (Kim et al., 2013), and reducing diet cost compared to other dietary fat sources (Figure 1.7; Jacobsen, 2014). In 2014, the rapid spread of porcine epidemic diarrhea virus across the U.S. resulted in a substantial increase in mortality (Hill et al., 2014). The concern of the perceived possibility of disease transmission through animal-derived feed ingredients prompted producers, nutritionists, and veterinarians to reevaluate the use animal-derived

Table 1.8. Fatty acid composition metabolizable energy content of animal, plant, and animal-vegetable blend lipid sources

		Fatty acids, % of total fat[1]										
Fat source	ME, kcal/kg	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	C20:1	C20:4	C22:5	C22:6
Beef tallow	7,835	3.7	24.9	4.2	18.9	36	3.1	0.6	0.3	0.00	0.0	0.0
Choice white grease	8,124	1.9	21.5	5.7	14.9	41.1	11.6	0.4	1.8	0.00	0.0	0.0
Poultry fat	8,364	0.9	21.6	5.7	6.0	37.4	19.5	1.0	1.1	0.10	0.0	0.0
Corn oil	8,579	0.0	10.6	0.1	1.9	27.3	53.5	1.16	0.1	0.00	0.0	0.0
Soybean oil	8,574	0.1	10.3	0.2	3.8	22.8	51.0	6.8	0.2	0.00	0.0	0.0
Animal-vegetable blend	8,225	1.5	20.2	3.2	10.1	35.5	21.6	0.9	0.6	0.03	0.0	0.0

[1] (NRC, 2012)

Figure 1.7. Cost of common dietary fat and oil sources including distiller's corn oil (DCO), soybean oil, yellow grease, and beef tallow used in swine growing-finishing pig diets.



Adapted from (Jacobsen, 2014)

feed ingredients in swine diets. Some pork producers have adopted precautionary steps by avoiding the use of animal fat sources, such as choice white grease, in swine diets and instead are electing to use plant-based oil sources, such as corn oil (Shoen, 2014).

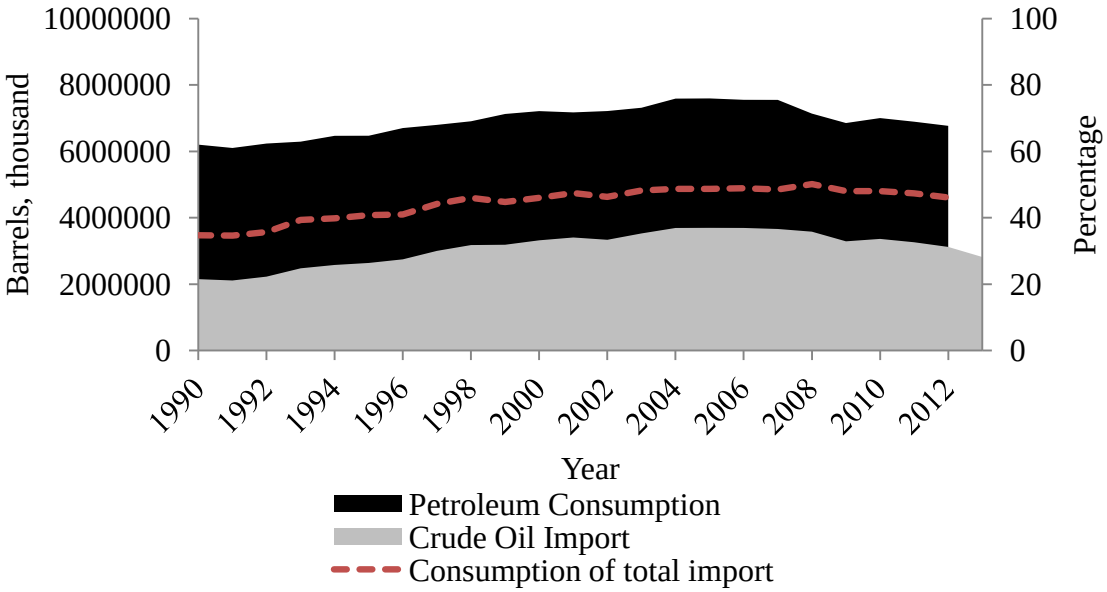
The increased use of corn oil is also due to the relatively low cost of distillers corn oil. The widespread adoption of technology to extract corn oil from thin stillage in ethanol plants has resulted in an increased supply of distillers corn oil, which has also become a cost effective alternative to feeding other dietary lipid sources (Figure 1.7; Jacobsen, 2014). The unsaturated fatty acids of corn oil are also more digestible than other more saturated fatty acid sources (Azain, 2001). Interestingly, the fatty acid digestibility of extracted corn oil, such as distillers corn oil, is greater than the fatty acid digestibility of corn oil in an 'intact form' present in DDGS (Kil et al., 2010). The primary limitation to increased dietary consumption of linoleic acid is the suppression of *de novo* lipogenesis in pig adipose tissue (Kouba and Mourot, 1998). As a result, the negative influence of unsaturated dietary fatty acids on fatty acid composition of pork fat is increased. This is of particular concern when feeding ingredients such as DDGS.

VII. Benefits and limitations of feeding DDGS diets to growing-finishing pigs

A. U.S. ethanol and DDGS production

The U.S. has historically used crude oil as the primary fuel source, relying heavily on imports (peaking at 3.7 billion barrels of crude oil annually in 2004 and 2005) to meet domestic demand (Figure 1.8; EIA, 2014a). Increased concern regarding the impact of burning fossil fuels on greenhouse gas emissions, climate change, along with the desire to become energy independent, prompted the U.S. Congress to pass the Energy Policy Act of 2005 to accelerate the blending of alternative and renewable fuels with gasoline

Figure 1.8. Volume of crude oil imported and petroleum consumed in the U.S. and percentage of consumption imported



Adapted from (EIA, 2014a)

(RFA, 2014c). Fuel ethanol was produced in significant quantities in the U.S. before 2005, but the Energy Policy Act of 2005 and the subsequent Energy Dependence and Security Act of 2007, set production quotas for production of biofuels under the Renewable Fuel Standard (**RFS**) to include: conventional, advanced, cellulosic, biomass, and undifferentiated advanced biofuels (RFA, 2014c).

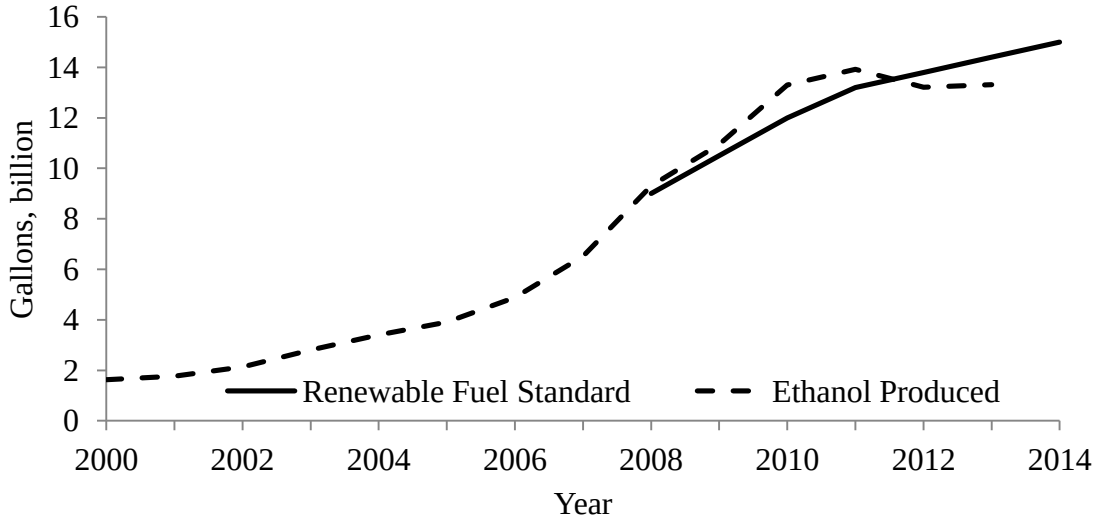
Ethanol produced from grain is considered a conventional biofuel (RFA, 2014c). Grains including corn, wheat, sorghum, and barley can be used as feedstocks for ethanol production (Liu, 2012). Corn is the predominant grain used in U.S. ethanol production because of its relative abundance, low cost, and high starch content compared with other grains (NRC, 2012; USDA, 2014a). Ethanol production subsidies were established to promote ethanol production which increased the demand for corn, and subsequently increased profit margins of corn production, thus encouraging farmers to plant more acres of corn. The last mandated subsidy increase for conventional biofuels is scheduled to

occur in 2015, which will total 15 billion gallons of ethanol (Figure 1.9; RFA, 2014c). These government policies, along with high crude oil prices (more than $100/barrel) in 2008 (EIA, 2014b), were the impetus needed to accelerate growth of the U.S. ethanol industry.

Ethanol production creates greater demand for corn, and increases the competition of corn for other uses, such as livestock and poultry feeds. In the U.S., corn has traditionally served as the primary energy source in swine diets (Woyengo et al., 2014). In 2013, 29.6% of total U.S. corn production was used for fuel ethanol production and 40.3% was used for livestock feed (NCGA, 2014). The increased demand for corn, along with suboptimal corn growing conditions in recent years, has caused record high corn prices, causing corn to become an expensive feed ingredient and dramatically increased pork production costs (Woyengo et al., 2014).

Due to growth of the U.S. ethanol industry, corn co-product supplies have also increased substantially, causing these co-products to become more available and

Figure 1.9. Annual U.S. ethanol production and Renewable Fuel Standard mandate

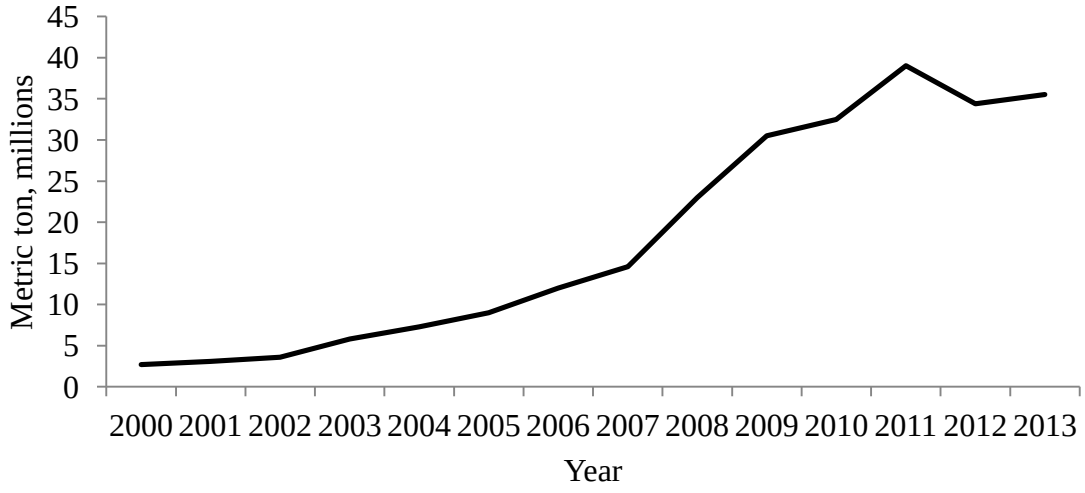


Adapted from RFA, 2014c

economical compared with more expensive feed ingredients, such as corn and soybean meal. Dried distillers grains with solubles is the most widely produced corn ethanol co-product, and in 2013, the U.S. produced 35.5 million metric tons of DDGS (Figure 1.10). In the U.S., most distillers grains are consumed by beef and dairy cattle (79%) and swine (12%; RFA, 2014a).

Use of DDGS extends outside of the U.S. due to growing demand in export markets and the record high cost of competing feed ingredients. Over 9 million metric tons (25% of U.S. DDGS production) was exported in 2013, with China (34.2%) and Mexico (16.0%) being the primary DDGS export markets (USGC, 2014). Not only are

Figure 1.10. Annual U.S. production of dried distillers grains with solubles



Adapted from (RFA, 2014a)

DDGS a valuable feed ingredient to livestock and poultry producers, but DDGS also represented 27% of gross revenues for ethanol plants in 2013 (RFA, 2014a). Even though DDGS are widely used in commercial swine diets, there are several benefits and limitations as a result of ethanol production methods.

B. Corn ethanol production and the influence on co-product composition

Ethanol plants produce several corn co-products from the non-fermented corn residuals that are suitable for livestock feed (Stein and Shurson, 2009). Several steps of ethanol production determine the nutritional composition and physical properties of each co-product, and affect the use of corn ethanol co-products in swine diets (Rosentrater et al., 2012). The dry grind system is the predominate method used for ethanol production (Rosentrater et al., 2012). Corn is ground to decrease particle size, increasing the surface area and allowing for greater access to starch for fermentation (Rosentrater, 2012), resulting in increased ethanol yields (Naidu et al., 2007). The cooking and liquefaction steps prepare the corn slurry for fermentation, with the primary purpose of gelatinizing starch granules and hydrolyzing glucose to shorter chain maltodextrins (Rosentrater et al., 2012). Additions of the enzymes α-amylase and gluco-amylase cleave the α-1,4 glycosidic and α-1,6 glycosidic bonds of the starch molecules. Ammonia (NH_3), lime ($CaCo_3$), or sulfuric acid (H_2SO_4) are added to adjust the slurry pH (Rosentrater et al., 2012). Yeast is added to ferment sugars resulting in the production of ethanol and carbon dioxide (Rosentrater et al., 2012).

Multiple steps throughout the corn ethanol production process elevate temperatures of co-product streams, but temperatures and duration of heating vary among ethanol plants and affect the nutritional value of the resulting feed co-products. Corn starch is fermented during ethanol production, and the residual non-fermented stream that does not proceed to distillation is called whole stillage (Rosentrater et al., 2012). The 2 major co-products yielded from whole stillage centrifugation are thin stillage and wet cake (Rosentrater et al., 2012). Through combinations of blending and centrifugation

steps, corn condensed distillers solubles (**CDS**) is produced from the thin stillage fraction and marketed to cattle feeders, or blended with distillers wet grains and sold for use in dairy or beef cattle rations, or the combination is dried to produce distillers dried grains with solubles (Rosentrater et al., 2012). More recently, about 75% of dry grind ethanol plants have adopted back-end oil extraction (RFA, 2014b) from the thin stillage fraction (USGC, 2012). The extracted corn oil can be used for biodiesel production or animal feed (RFA, 2014c).

While data are limited, the feeding of 15% corn distillers solubles to growing-finishing pigs has resulted in reduced feed intake and average daily gain compared to feeding a corn-soybean meal diet (Squire et al., 2005). Feeding diets containing wet distillers co-products (e.g. 20% wet distillers solubles, 30% wet distillers grains, or 25.5% wet distillers grains with 4.5% wet distillers solubles) did not compromise growth performance compared to feeding corn-soybean meal diets or diets with dried distillers grains with solubles (Badioo et al., 2014). Marketing these wet co-products reduce cost of production for ethanol plants because they are not dried, but the high moisture content of corn ethanol wet coproducts increases the transportation cost and reduces shelf-life (Cao et al., 2009). Swine farms would need to be located close to ethanol plants to make the feeding of wet co-products feasible (DeRouchey and Richert, 2010). Corn distillers solubles and wet distillers grains with solubles are best suited for use in liquid feeding systems, which are not widely used in the U.S (DeRouchey and Richert, 2010). Therefore, corn distillers solubles and wet distillers grains with solubles are not suitable ingredients to use in swine diets.

Corn DDGS are 3 times higher in fiber, protein, lipid, and minerals content than corn (Stein, 2012). However, the nutrient content of DDGS is highly variable between (Han and Liu, 2010) and within plants due to the adoption of new production technologies, variations in processing techniques (drying temperature and time), oil extraction, fermentation efficiency (Liu, 2011), and the addition rate of CDS (Cao et al., 2009). On a dry matter basis, the corn oil concentration increases from the whole stillage to thin stillage stage, with the resulting CDS having a greater concentration of oil compared to distiller grains (Han and Liu, 2010). Thus, the oil content of the resulting DDGS is intermediate to the concentration in thin stillage and distiller grains on a dry matter basis (Han and Liu, 2010). This also means that the proportion of thin stillage or CDS that is added to the wet cake increases crude fat of DDGS (Cao et al., 2009).

C. Advantages and disadvantages of DDGS in swine diets

The energy value of feed ingredients comes from carbohydrates, lipids, and protein (Patience, 2012). Each nutritional component of DDGS provides advantages and limitations when fed to pigs. Several nutritional characteristics of DDGS make it a well-suited feed ingredient for swine diets including: similar metabolizable energy content to corn (NRC, 2012), relatively high crude protein and lysine content (Almeida et al., 2011), high phosphorus content (NRC, 2012), and high phosphorus digestibility (Almeida and Stein, 2010). However, carcass dressing percentage can be reduced when adding DDGS to the diet due to its high fiber content (Graham et al., 2014; Xu et al., 2010b). Furthermore, increasing dietary levels of DDGS reduces pork fat firmness due to the

Table 1.9. Energy and nutrient composition of corn ethanol co-products adjusted to 100% dry matter basis[1,2]

				Corn dried distillers grains with solubles		
	Corn	CDS[3]	DDG[4]	> 10% oil	6-9% oil	< 4% oil
Gross energy, kcal/kg	4,454	5,372	5,416	5,429	5,271	5,712
Metabolizable energy, kcal/kg	3,858	3,198	3,477	3,845	3,801	3,476
Net energy, kcal/kg	3,036	2,312	2,322	2,669	2,622	2,251
Crude protein, %	9.3	21.30	31.81	30.60	30.62	31.22
Total lysine, %	0.28	0.91	1.20	0.86	1.01	0.76
SID lysine[5], %	0.21	NA	0.76	0.53	0.61	0.47
Crude fat, %	3.9	13.75	9.57	11.68	9.96	4.00
Neutral detergent fiber, %	10.3	28.25	46.09	36.39	34.09	37.82
Acid detergent fiber, %	2.8	8.54	17.12	13.16	13.45	18.95
Calcium, %	0.02	0.33	0.09	0.13	0.09	0.06
Phosphorus, %	0.29	1.41	0.62	0.82	0.67	0.85
Sulfur, %	NA	0.42	NA	0.74	0.54	NA
Sodium, %	0.02	0.30	0.10	0.25	0.34	NA

[1]Adapted from (NRC, 2012)
[2] NA = Not available
[3] CDS = Corn distillers solubles
[4] DDG = Dried distillers grains
[5] SID = standardized ileal digestibility

high linoleic acid content in corn oil present in DDGS (Cromwell et al., 2011; Xu et al., 2010b).

1. Energy value of DDGS

Energy is the most expensive component of swine diets (Patience, 2012). The gross, digestible, and metabolizable energy content of DDGS is equal to or superior to corn (Stein and Shurson, 2009). However, the net energy content of DDGS is lower than that of corn (NRC, 2012) because of its relatively high crude protein and fiber content, and low starch content. Crude protein and fiber are utilized less efficiently as energy sources than energy from starch and crude fat because more energy from crude protein and fiber 'escapes' as heat and cannot be used by the pig for growth (Noblet and van Milgen, 2013). As a result, the high crude protein and fiber content of DDGS limit the use of DDGS in swine diets (Stein and Shurson, 2009).

2. Crude protein and lysine content in DDGS

Even though DDGS has a greater lysine content than corn, the standardized ileal digestibility of lysine of DDGS is poorer than corn (69.2 vs. 46.0%; Almeida et al., 2011). However, the standardized ileal digestibility of lysine in DDGS is quite variable and can range among DDGS sources from 51.4 to 74.5% (Pahm et al., 2008). Furthermore, the standardized ileal digestibility of lysine of DDGS is lower than DDG (77.9%; Pahm et al., 2008). Thus, the reduced standard ileal digestibility of lysine in DDGS is attributed to the addition of CDS. Furthermore, the variability in lysine digestibility of DDGS is affected by amount of CDS added to the wet grains before drying to produce DDGS (Curry et al., 2014). Increasing the proportion of CDS in DDGS likely increases the need for further drying of DDGS (Liu, 2011). Lysine is particularly

sensitive to functional degradation by heat because of its chemical structure. After peptide bonds are formed between an amino and carboxyl group, lysine has an additional amino group, that in the presence of sugars, condenses and when heated leads to a Maillard reaction (Ames, 1998). Several heating steps occur throughout the ethanol production process such as during the jet cooker, saccharification, and co-product drying steps (Rosentrater et al., 2012). Lower efficiency of starch fermentation could also contribute to increased sugars in the CDS (Naidu et al., 2007), leading to greater potential for sugar-amino group reactions and thus, decrease lysine digestibility of DDGS.

The protein quality of DDGS is poor due to the high crude protein content relative to the lysine content. This can result in greater nitrogen intake and excretion of non-utilized nitrogen compared to feeding a corn-soybean meal diet (Liu et al., 2012). Deamination of amino groups from excess amino acids increases energy utilization and nitrogen excretion resulting in these amino acids being unavailable for use in lean growth (van Milgen et al., 2001). However, use of the net energy system accounts for the energy lost as heat increment during these processes (Noblet and van Milgen, 2013).

Nitrogen content of manure is an important consideration when manure is applied to cropland as a fertilizer because excess nitrogen can leach into water supplies (Fan, 2013). Poor protein quality in DDGS can be overcome by adding supplemental amino acids to swine diets (Stein and Shurson, 2009). Immunologically castrated are likely to be more sensitive to dietary imbalances in amino acids and lower lysine digestibility of diets containing DDGS because they have greater digestible amino acid requirements compared to physically castrated pigs.

3. Fiber in DDGS

Dried distillers grains with solubles has a relatively high content of fiber (NRC, 2012), and structural carbohydrates, such as fiber, are poorly utilized by the pig (Urriola et al., 2013a). Some energy can be captured through fiber fermentation in the hindgut and production of volatile fatty acids (Kerr and Shurson, 2013). However, like crude protein, fiber digestion generates a large heat increment compared to other nutrients such as starch and lipid, use of the net energy system accounts for differences in heat increment. Thus, dried distillers grains with solubles has a lower net energy compared to corn. Since fiber has not traditionally been viewed as an attractive option to feed to pigs, fiber is poorly understood (Kerr and Shurson, 2013). Rapid and low-cost analytical methods are being developed to determine *in vitro* digestibility of high-fiber feed ingredients (Huang et al., 2014). Given the general high cost of feed ingredients, implementing technology to enhance energy utilization from fiber would be advantageous to capture more of the energy from high fiber feed ingredients, such as DDGS.

An additional drawback to feeding DDGS to growing-finishing pigs is its effect on reducing carcass dressing percentage, which has been attributed to fiber increasing visceral mass. Feeding diets containing 30% DDGS for 28 days increased the percentage of portal-drained visceral mass (sum of spleen, pancreas, stomach, cecum, small intestine, colon and rectum) by 9.4% compared to feeding a corn-soybean meal diet (Agyekum et al., 2012). The viscera accounts for a large portion of whole-body energy expenditure (de Lange et al., 2001). Therefore, from a growth and nutrient utilization perspective, increased visceral mass reduces the energy available for lean growth. Feeding strategies to overcome reduced carcass dressing percentage will be discussed later in this review.

The high fiber content of DDGS also causes manure management challenges. The decreased dry matter digestibility of DDGS results in greater manure volume (Stein and Shurson, 2009) than when feeding corn-soybean meal diets. The increased fermentation of complex carbohydrates also increases methane production (Jarret et al., 2011). Greater methane production and increased dry matter in manure have also been anecdotally implicated, along with the lipid content of DDGS, to cause manure foaming in anaerobic pits, resulting in serious animal welfare and worker safety concerns due to barn explosions and flash fires (Yan, 2014). Stable foam formation requires gas, a surfactant, and a stabilizer (Jacobson et al., 2013). These characteristics have been theoretically linked to feeding DDGS diets, where methane is produced from fiber fermentation, the lipid content of DDGS serves as a surfactant, and the undigested dry matter and filamentous bacteria serves as a foam stabilizer (Jacobson et al., 2013). Recent research has determined that decreasing diet particle size from 631 to 374 microns improved dry matter, lipid, and fiber digestibility which also reduced manure foaming capability (unpublished data). Interestingly feeding 21% soybean hulls resulted in greater manure foaming capability compared to feeding 35% DDGS (unpublished data). Currently, there is no conslusive evidence that feeding DDGS diets increases manure foaming capability. Further understanding of the dynamics of manure pit foaming are necessary to identify causes and solutions that can overcome manure foaming.

4. Lipids in DDGS

Previous reports indicate traditional DDGS contains 10.8 to 12.0% crude fat (Belyea et al., 2010), and Pedersen et al (2007) reported crude fat percentage to range from 9.59 and 14.25% compared to a corn reference with 3.34% crude fat, adjusted to dry

basis. Within DDGS sources, the coefficient of variation percentage has been reported to range from 7.4 to 19.1% (Belyea et al., 2010), while Spiehs et al., 2002) reported crude fat coefficient of variation percentage ranging from 4.4 to 10.5%. More recently, ethanol manufacturers have recognized the additional revenue, with minimal capital investment, that can be obtained by marketing corn oil as an additional co-product. As a result, at least 75% of ethanol manufactures are extracting a portion of corn oil from the thin stillage by centrifugation (RFA, 2014b), and subsequently producing reduced oil DDGS which contains 7 to 9% crude fat (USGC, 2012). Oil extraction causes compositional changes and increases variation in energy and nutrient content of DDGS. In fact, the NRC (2012) has expanded the ingredient composition section to now specify three different compositional profiles of DDGS based on corn oil content of (> 10%, 5 to 9%, and < 5% ether extract; Table 1.7). It has been assumed that the high metabolizable energy content of DDGS is primarily due to the high concentration of corn oil, which contains about 2.25 times more energy than carbohydrates (Kerr et al., 2013). As a result, oil extraction was expected to reduce the metabolizable energy value of DDGS in swine diets. However, lipid content of DDGS has been shown to be a poor indicator of the metabolizable energy content of DDGS with variable oil content (Kerr et al., 2013). Additionally, reduced oil DDGS (7.5 and 6.9% lipid content) has been shown to have reduce standardized ileal digestibility of amino acids compared to ”traditional” DDGS (containing 11.5% oil; Curry et al., 2014). The amino acid digestibility differences reported by Curry et al. (2014) can be explained by the higher dietary lipid content found in “traditional DDGS” reducing gastric emptying, which in turn slows the rate of digesta passage allowing for increased time for peptide and amino acid digestion, and thus,

resulting in greater amino acid digestibility (Curry et al., 2014). The continually evolving processes used in ethanol and DDGS production, substantially alter the composition and nutrient digestibility of the resulting DDGS. Consequently, this increase in variability in feeding value creates difficulty in determining accurate nutrient estimates to use in diet formulation with this ingredient. Inaccurate estimates for metabolizable or net energy and digestible amino acid content of various DDGS sources can lead to inaccurate diet formulations and suboptimal growth performance.

5. Managing energy and nutrient variability among DDGS sources for accurate swine diet formulation

Use of near-infrared spectroscopy is becoming widely used in the feed industry to provide nutritionists with a fast and reliable method to estimate nutrient composition of feed ingredients (Graham et al., 2013). This method requires a large database to build robust calibrations using wet chemistry composition (e.g. CP, fat, starch). Chemical composition of DDGS have been used as inputs for gross, digestible, and metabolizable energy to develop prediction equations (Anderson et al., 2012; Kerr et al., 2013; Pedersen et al., 2007; INRA, 2008). These equations were cross-validated with previously published nutrient composition data for various sources of DDGS and *in vivo* DE and ME estimates to determine that the Anderson et al. (2012) prediction equation had the highest accuracy and precision (Urriola et al., 2014). The Anderson et al. (2012) ME equation was further validation by formulating diets using different DDGS sources that had similar predicted metabolizable energy content, but contained oil content of 5.87, 9.85, or 14.23% (Wu et al., 2014). The ME content of DDGS with 9.85 and 14.23% oil was accurately predicted using the Anderson et al. (2012) equations (Wu et al., 2014).

However, ME content of DDGS with 5.87% oil may have been under or over estimated by the Anderson et al. (2012) equation. Over estimation could be due to fewer DDGS sources representing a lower lipid content when the equations were developed (Wu et al., 2014). However, this equation could have under estimated the ME value of DDGS with 5.87% oil due to overestimation of ME from fat or CP, or unaccounted digestibility of nutrients. Low-oil DDGS may have placed greater pressure on the accuracies of CP and NDF estimates in the ME equation by Anderson et al. (2012). As more studies evaluating the chemical composition and *in vivo* ME content of low oil DDGS sources are conducted, it may be possible to improve the accuracy of equations for predicting ME content of low oil DDGS. Improving the precision and accuracy of energy prediction equations will allow nutritionists to manage variability in metabolizable energy content of reduced oil DDGS for more accurate swine diet formulations.

6. Phosphorus content and digestibility

The phosphorus digestibility of corn is relatively poor because most of the phosphorus is present as phytate (Kornegay, 2001). In a corn-soybean meal based diet, the phosphorus requirement of pigs can be met by supplementing diets with inorganic phosphorus (Baker and Stein, 2013). Phosphorus that is not used by the pig, including undigested phytate phosphorus, is excreted in feces (NRC, 2005). Excretion of undigested phosphorus is not cost effective because phosphorus is the third most expensive nutritional component in swine diets (NRC, 2005). Excretion of undigested phosphorus also causes concerns related to environmental sustainability of pork production systems (NRC, 2005). Phosphorus in swine manure has a valuable role in crop production as a fertilizer, but application rates are limited by soil phosphorus content

(NRC, 2005). Phosphorus remains in the topsoil making it vulnerable for movement to surface waters as a result of soil run-off, which can lead to eutrophiciation (Fan, 2013). During fermentation of corn during ethanol production, phytate bonds are hydrolyzed by yeast phytase which improves digestibility of phosphorus in DDGS (Liu, 2011). The phosphorus content and standardized total tract digestibility of DDGS is greater than corn (0.82 vs. 76.5% and 0.19% vs. 42.5%, respectively; Rojas et al., 2013). Use of ingredients with greater phosphorus digestibility, such as DDGS, improves the nutritional efficiency of pork production, reduces the cost of swine diets, and is more environmentally sustainable.

7. Mycotoxins

Mycotoxins are toxins produced from fungi produced in corn (Bryden, 2012). When present, mycotoxins can be a major limitation for feeding DDGS to pigs (Stein, 2012). There are many fungal species, but toxins of greatest concern in pork production are aflatoxins, deoxynivalenol, fumonisins, zearalenone, and ochratoxin (NRC, 2012). In corn, mycotoxins are located in the germ and pericap portions of the kernel (Liu, 2011). Since the germ and pericap contain low levels of starch (Liu, 2011) they are not fermented, thus mycotoxins remain in the whole stillage portion and are concentrated by a factor of 3 times the concentration in corn (Liu, 2011). Growth of these fungal species and the production of toxins is dependent on many factors including environment conditions during growing, harvesting, and storage, so mycotoxin issues occur sporadically (Bryden, 2012). Different mycotoxins can be produced under vastly different conditions. For example, aflatoxins develop in hot, humid climatic conditions when plants are heat-stressed, while fusarium develops predominantly under cold, wet

conditions (Liu, 2011). It is recommended that grain be stored with moisture content less than 14% to avoid mycotoxin production during storage (Zhang and Caupert, 2011). In ethanol production, mycotoxins can decrease ethanol yields by placing stress on yeast used for starch fermentation (Liu, 2011). This is important not only due to the presence of mycotoxins in the resulting co-products, but lower ethanol yields would result in more unfermented sugars in the stillage, which could increase the likelihood of Maillard reactions that reduce the standardized ileal digestibility of lysine. The presence of mycotoxins in DDGS can lead to feed refusal, poor growth performance, and immunosuppression (NRC, 2012). Use of increasing levels of DDGS in swine diets can increase these negative effects when mycotoxins are present. Since immunological castration relies on antibody production of GnRF, the immunosuppressive effect of feeding mycotoxin contaminated feed may hinder antibody production. Thus, feeding mycotoxin contaminated feed to immunologically castrated pigs maybe an even greater risk than physically castrated pigs, but this possibility has not been evaluated.

8. Physical properties of DDGS

Physical properties of corn DDGS are variable due to variation of ethanol production procedures. Some of the poor handling and storage properties of DDGS are characterized by relatively low bulk density, as well as variable moisture content and particle size (Rosentrater, 2012). Dried distillers grains with solubles is a bulky feed ingredient limiting the mass of DDGS per unit of volume for storage or transportation, and increases the cost per unit storage and transportation of DDGS (Woyengo et al., 2014). Increasing the moisture content of DDGS and addition of greater proportions of CDS to DDG increases compressibility of DDGS, reducing flowability properties

(Ganesan et al., 2008). Dried distillers grains with solubles is also a hygroscropic material, and one study reported moisture of DDGS increased from 9.05% to 12.26% over a 13 week period in storage (USGC, 2012). In general, DDGS is considered a cohesive material and during the drying process, particles can agglomerate to create "syrup balls" (Rosentrater, 2012). Flowability properties of DDGS can be improved by controlling the amount of CDS added to distillers dried grains, as well as pelleting DDGS, which improves its bulk density (USGC, 2012). Complete diets formulated with 30% DDGS had decreased feed flowability compared to standard corn-soybean meal diets, and reducing the diet particle size of DDGS from 818 to 594 and 308 μm further reduced feed flowability (Liu et al., 2012). The beneficial trade-off of reducing particle size of DDGS is an increase in metabolizable energy from DDGS of 13.6 kcal/kg DM for every 25 μm decease (Liu et al., 2012). Oil extraction (from 9.3 to 2.1% dry matter basis) has also been shown to improve flowability of DDGS (Ganesan et al., 2009). Therefore, oil extraction from thin stillage prior to manufacturing DDGS should improve DDGS flowability.

D. Growth performance of growing-finishing pigs fed DDGS diets

Stein and Shurson (2009) conducted a literature summary including 25 experiments from North America, where pigs were fed diets containing up to 30% DDGS during the growing-finishing phase. A more recent summary of 21 experiments was compiled by (Hardman, 2014) and included experiments where diets containing up to 60% DDGS were fed. Of the experiments summarized, average daily feed intake remained unchanged in 65% (Stein and Shurson, 2009) and 62% (Hardman, 2014) of experiments, and decreased in 26% of experiments (Stein and Shurson, 2009). Gain

efficiency was unchanged in 64% (Stein and Shurson, 2009; Hardman, 2014) of experiments, and was reduced in 20% of experiments reviewed by Stein and Shurson, 2009). Average daily gain was unchanged in 72% (Stein and Shurson, 2009) and 67% (Hardman, 2014) of the experiments summarized, while ADG was reduced in 24% of experiments evaluated by Stein and Shurson, 2009). Inaccurate metabolizable energy and digestible amino acid values, along with nutrient variability among DDGS sources, may have resulted in reduced growth performance in some studies (Stein and Shurson, 2009). Nutritional variability of DDGS creates a challenge for accurately formulating swine diets, and this challenge has increased with the adoption of oil extraction technologies by ethanol plants. The price relationship between corn, soybean meal, and DDGS sometimes warrants using more than 30% DDGS in the diet to decrease cost. Few studies have been conducted to evaluate diets containing more than 30% DDGS.

A study was conducted involving nine universities, representing a variety of geographical and climate regions and a variety of management systems to determine the growth performance of pigs fed diets containing up to 45% DDGS (Cromwell et al., 2011). Diets were manufactured at each location and used locally available corn and soybean meal (Cromwell et al., 2011). However, the same source of DDGS was used among all locations. Increasing the dietary DDGS inclusion level up to 45% resulted in a linear decrease in overall ADG (Cromwell et al., 2011). These changes in ADG were the result of a cubic decrease of overall ADFI, where pigs fed diets containing 30 and 45% DDGS had less ADFI compared to pigs fed diets containing 0 and 15% DDGS (Cromwell et al., 2011). Overall G:F was different among dietary treatments among collaborating universities (Cromwell et al., 2011), but gain efficiency values for each

university were not reported. However, these results suggest that factors other than the DDGS source and DDGS nutrient variability may contribute to the variation in growth performance responses summarized by Stein and Shurson, 2009) and Hardman (2014). Other factors contributing to the variability in growth performance may be inherent to climate, management, or nutrient content of other ingredients, such as corn and soybean meal. For example, it is possible that pigs raised in hot climates may be more sensitive to high fiber diets, such as DDGS, than in cooler climates because of the greater heat increment from fiber consumption (Noblet and van Milgen, 2013).

In studies that involved feeding diets containing 60% DDGS, ADFI was not different between pigs fed diets containing 20 and 60% DDGS, but ADG was reduced in pigs fed 60% DDGS compared to pigs fed 20% DDGS diets due to poorer feed efficiency for pigs fed 60% compared to 20% DDGS diets (Bergstrom et al., 2009a). However, in a study were 30 and 60 % DDGS diets were fed, overall growth performance was not affected (Weber et al., 2013). The variable responses to growth performance when feeding diets containing up to 60% DDGS, especially the lack of a differences reported by Weber et al. (2013), could be due to the absence of using corn-soybean meal control diets in these studies . Compared to other studies, pigs fed 30 and 60% DDGS in the Weber et al. (2013) study had lower ADFI than pigs in the Bergstrom et al. (2009a) and Cromwell et al. (2011) studies. Hardman et al., (2014) also fed diets containing up to 60% DDGS (including a corn-soybean meal control) and observed a linear decrease in ADG and ADFI, but no difference in gain efficiency. In general, it appears that studies that contain diets with more than 30% DDGS are more likely to result in reduced ADFI than studies feeding where diets containing 30% DDGS or less were fed. This may be a

result of pigs having physical gut capacity limitations due to the bulkiness of fiber in diets containing high amounts of DDGS. This also emphasizes the need to obtain accurate energy and nutrient estimates to achieve adequate nutrient intake with increasing levels of DDGS. The effect of variable DDGS nutrient composition may be magnified when feeding immunologically castrated because they have a higher demand for lean growth and thus need sufficient amino acid and energy intake to achieve optimal lean gain. Additionally, it is unknown if the physical gut fill limitation related to high fiber diets would impede the expected increase of ADFI after the second Improvest® dose.

E. Carcass dressing percentage of pigs fed DDGS diets

A literature summary of studies reported by Stein and Shurson (2009) included studies where feeding diets containing up to 30% DDGS showed that in 44 and 56% of studies, dressing percentage decreased or was unchanged, respectively. A linear decrease in dressing percentage has been observed when pigs were fed diets containing up to 30% (Xu et al., 2010b; Whitney et al., 2006), 40% (Graham et al., 2014), and 60% DDGS (Hardman, 2014; Leick et al., 2010), but this did not occur when Xu et al. (2010a) and Cromwell et al. (2011) fed diets containing up to 30 or 45% DDGS, respectively. When feeding diets containing 30% DDGS, it has been observed that the portal-drained visceral mass increases (Agyekum et al., 2012). Asmus et al., (2014b) observed that the full large intestine, large intestine with digesta contents removed, and rinsed large intestine of pigs fed 30% DDGS and 19% wheat midds diets for 90 days was 25%, 15%, and 12% greater, respectively, than pigs fed corn-soybean meal diets (Asmus et al., 2014a). While not reported, the authors implied that digesta mass was also greater in pigs fed diets with 30% DDGS plus 19% wheat middlings (Asmus et al., 2014a), which is logical due to the

bulkiness and water retention capability of fiber (Urriola et al., 2013a). The discrepancy of dressing percentage results between Xu et al. (2010a) and Cromwell et al. (2011) could be related to ADFI and gut fill immediately prior to harvest. Cromwell et al. (2011) did not observe any difference in ADFI in the final dietary phase when feeding up to 45% DDGS, and Xu et al. (2010a) only reported overall ADFI, but did not observe in any difference in ADFI when feeding up to 30% DDGS.

F. Lean quality from feeding DDGS diets

Lean quality generally receives less attention than fat quality when feeding DDGS to pigs because of the minimal lipid content of lean (Apple et al., 2009c), and for pork loins (IMPS #410), subcutaneous fat is trimmed to at least 0.10 cm (IMPS, 2014). Results from a study that measured color of the longissimus muscle from pigs fed up to 30% DDGS showed a linear decrease in Minolta a* and b* values (Xu et al., 2010b). The linear decrease in b* reported by Xu et al. (2010) is consistent with others who have also reported a linear decrease in b* when pigs were fed diets with up to 20% DDGS (Moreno et al., 2010; Widmer et al., 2008). In contrast, feeding diets containing up to 60% DDGS did not result in any differences in L*, a*, or b* of the longissimus muscle (Leick et al., 2010). Corn contains yellow pigments called carotenoids (Hugo and Roodt, 2007). Since these are lipids, and thus, are not fermented during ethanol production, they remain in the co-products in a concentrated form. However, due to the drying processes of ethanol and DDGS production it is estimated that up to 50% of these pigments may be destroyed (Winkler-Moser, 2012). The variability of drying processes among ethanol plants used to manufacture DDGS, along with variability in carotenoid content, may result in inconsistent objective b* values of the longissimus muscle.

Subjective color, marbling, and firmness scores were unaffected when up to 20 (Widmer et al., 2008) or 30% DDGS was included in swine diets (Whitney et al., 2006; Xu et al., 2010b). No difference in subjective color score was observed. However, subjective firmness and marbling linearly decreased when pigs were fed up to 30 (Xu et al., 2010b) and 60% DDGS diets (Leick et al., 2010). The reduced subjective marbling scores could also explain the reduced firmness scores because less intramuscular fat would suggest a lower overall proportion of triglyercides and higher proportion of phospholipids in the longissimus muscle. The longissimus muscle has a greater proportion of phospholipids relative to triglycerides compared to subcutaneous backfat. The phospholipid fraction has a much greater unsaturated fatty acid profile, and contains 2.5 times more linoleic acid and 50 times more arachidonic acid than the triglyceride fraction (Wood et al., 2008). Since the melting point of fatty acids is lower with increasing concentration of unsaturated fatty acids (Azain, 2001), this could also contribute to the reduced firmness in loins.

Polyunsaturated fatty acids are also more susceptible to peroxidation due to the lower energy needed to abstract hydrogens from interrupted methylenes of the carbon chain (Wood et al., 2008). Thus, lean can also be susceptible to lipid peroxidation due to the presence of polyunsaturated fatty acids. Thiobarbituric acid reactive substances **TBARS**), measured as malonaldehyde equivalents, is a commonly used measure to quantify the peroxidation of polyunsaturated fatty acids (Shahidi and Wanasundara, 2008b). Feeding diets with up to 30% DDGS did not increase lipid peroxidation as measured by TBARS (Xu et al., 2010b), but greater dietary amounts of DDGS (45 and

60%) increased TBARS after 21 days of storage, suggesting more lipid peroxidation than when feeding diets in excess of 30% DDGS (Leick et al., 2010).

Reactive oxygen species produced in antemortem can cause premature release of calcium from the sarcoplasmic reticulum in early postmortem, accelerating pH decline, and impairing water-holding capacity (Barbut et al., 2008). Decreased water holding capacity of lean is an undesirable quality characteristic of pork, and was observed when feeding up to 60% DDGS (Leick et al., 2010). While data are limited, it appears that feeding growing-finishing pigs diets with more than 30% DDGS has greater negative effects on lean quality than feeding diets containing less than 30%. More attention needs to be given to lean quality and the polyunsaturated fatty acid content of lean because this could be related to the lower subjective firmness scores, greater TBARS, and decreased water-holding capacity of pork loins.

G. Pork fat quality of pigs fed DDGS

1. Pork fat color

Today, pork fat quality preferences span across geographical regions where Japanese markets have a preference for firm, bright white pork fat (Morgan et al., 1995) compared to poor fat quality that would be described as wet and grey (Hugo and Roodt, 2007). Japan is the largest export market for U.S. pork based on product value (USMEF, 2013). Therefore, maintaining acceptable pork fat color is essential for maintaining and increasing demand for U.S. pork in Japan.

Fat color can be measured subjectively, using the 4-point Japanese Color Score scale where 1 = bright white and 4 = yellow, or objectively with a colorimeter. These color measurements are limited by the fact that there is no known hedonic scale

associated with these color scales. Feeding diets containing up to 30% DDGS did not change objective Minolta belly fat color (L*, a*, b*) or subjective Japanese Color Score, but backfat L* decreased and b* increased (Xu et al., 2010b). Pigs fed 20 and 30% DDGS diets had lower backfat L* values compared with pigs fed corn-soybean meal diet (Xu et al., 2010b). The increasing dietary levels of DDGS up to 60% resulted in a linear decrease in belly fat Minolta L* value, but no change in a* or b* values (Leick et al., 2010). Changes in fat color due to feeding DDGS are inconsistent, but darker (lower L*) and more yellow (higher b*) carcass fat might be related to carcass fat thickness because pigs with less carcass fat have a greater proportion of connective tissue compared to fatter carcasses (Hugo and Roodt, 2007). As with lean color, the variation among studies assessing fat color could be related to variation of carotenoid activity of the DDGS source fed, and any negative effects would be reduced with oil extraction. The inconsistency in color scores among fat depots is peculiar, but since fatty acid composition of carcass fat depots changes disproportionally among depots when feeding DDGS, one hypothesis is that carotenoids are preferentially deposited with specific fatty acids.

2. Fatty acid composition and iodine value

Methods of assessing pork fat quality and composition differences among depots have been previously discussed, where fatty acid composition and iodine value are the most universally used methods of assessing pork fat quality. A summary of peer-reviewed research identified that 7 out of studies 8 observed an increase in belly fat iodine values, and the remaining study observed no change in belly fat iodine value when feeding growing-finishing pigs diets containing up to 30% DDGS (Stein and Shurson, 2009). A more recent review by Hardman, 2014) identified 1 out of 9 experiment where

iodine value of belly fat was not different. In that study, pigs were fed diets containing 0, 10, or 20% DDGS. In all other studies, the maximum dietary DDGS inclusion level ranged from 30 to 60% and resulted in increased belly fat, backfat, and jowl fat iodine value with increasing DDGS level in the diet (Hardman, 2014). This is due to the profound effect that feeding DDGS has on altering the fatty acid composition, particularly linoleic acid content of pork fat depots. Linoleic acid content of pork fat depots is usually increased 2-fold by feeding diets containing 30% DDGS compared to corn-soybean meal control diets (Xu et al., 2010a; Xu et al., 2010b; Cromwell et al., 2011). Iodine value, calculated using either the AOCS (1998) or Meadus et al. (2010) equations, is greatly influenced by the linoleic acid content of pork fat. The negative effects of feeding DDGS on carcass dressing percentage and reduced fat firmness can be overcome by using specific feed formulation and feeding strategies.

VIII. Feeding and feed formulation strategies to overcome reduced pork fat quality and carcass dressing percentage when feeding DDGS diets to growing-finishing barrows and gilts

A. Carcass dressing percentage

As previously described, feeding DDGS to growing-finishing pigs can, but does not always, decrease carcass dressing percentage. Several studies have been conducted to evaluate the effect of removing the DDGS before harvest and feeding a standard corn-soybean diet. In a study where 30% DDGS diets were fed, carcass dressing percentage was reduced compared to feeding a corn soybean meal diet, but was improved by removing the DDGS from the diet for 3 or 6 weeks before harvest (Gaines et al., 2007). However, a 6 week withdrawal was required to fully restore carcass dressing percentage

similar to that of pigs fed a standard corn-soybean meal diet (Gaines et al., 2007). It has also been shown that increasing the withdrawal period from diets with 30% DDGS and 19% wheat middlings to linearly improve carcass dressing percentage (Coble et al., 2013). However, some have observed that withdrawing 30% DDGS from the diet for 30 days (Hill et al., 2008), 20 or 40 days (Jacela et al., 2009), or 21, 42, 63 days (Xu et al., 2010a) before harvest was ineffective for improving carcass dressing percentage. In these studies, there was no difference in carcass dressing percentage between pigs fed corn-soybean meal vs. 30% DDGS diets because there was no margin for improving in carcass dressing percentage. Using a withdrawal feeding strategy can be effective in improving carcass dressing percentage when dressing percentage decreases due to feeding high fiber diets.

B. Pork fat quality

1. Feeding strategies

The inherent differences in fatty acid composition of dietary fat sources directly alters the fatty acid composition of pork fat depots (Apple et al., 2009a). Several feeding strategies have been developed that focus on controlling the intake of polyunsaturated fatty acids in diets of growing-finishing pigs. The use of corn germ, beef tallow, palm kernel oil, and glycerol in growing-finishing diets has been found to improve fat firmness when serving as the sole source of supplemental lipid to the diets (Lee et al., 2013). Lee et al. (2013) investigated the effects of adding these lipid sources to diets containing 30% DDGS as possible corrective measures to improve pork fat quality. Unfortunately, these sources and dietary inclusion levels (corn germ = 15%, beef tallow = 3%; palm kernel oil = 3%; glycerol = 5%) did not improve fat quality (Lee et al., 2013). Interestingly, fatty

acid composition of backfat and belly fat from pigs fed DDGS or DDGS with corn germ, beef tallow, palm kernal oil, or glycerol were not different from pigs fed a standard corn-soybean meal diet and resulted in similar backfat and belly fat IV across all dietary treatments (Lee et al., 2013). Results from Lee et al. (2013) were likely influenced by the unusually high linoleic acid content of back and belly fat of pigs fed corn-soybean diets (20.4% and 18.69% in backfat and belly fat, respectively). Typically, carcass fat linoleic acid content of pigs fed corn-soybean meal diets ranges from 9.12 (Xu et al., 2010b) to 13.56% (Cromwell et al., 2011) in backfat and from 9.19 (Xu et al., 2010a) to 11.63% (Asmus et al., 2014b) in belly fat. Others have reported that adding 5% tallow in addition to feeding pigs 30% DDGS reduced belly fat iodine value but not backfat iodine value (Pomerenke et al., 2011). In general, adding more saturated fatty acid sources to diets containing high concentrations of unsaturated fatty acids (i.e. DDGS) is ineffective for improving carcass fat iodine value, but other factors such as the length of feeding each lipid source may also play a role in the ability to improve pork fat quality.

Increasing the feeding duration of diets containing yellow grease resulted in a quadratic increase in linoleic and linolenic acid content in jowl fat, while increasing the feeding duration of tallow resulted in a quadratic decrease in linoleic and linolenic acid in jowl fat (Browne et al., 2013). By switching fat sources from yellow grease to tallow in the last two dietary phases, Brown et al. (2013) observed that backfat, but not jowl fat, iodine value was restored to levels similar to when tallow was fed throughout the growing-finishing period. In general, unsaturated pork fat is easily created but much more difficult to reverse. Several factors need to be considered including dietary lipid and fatty acid intake and length of feeding dietary lipid sources. With these factors in mind, other

feeding strategies and changes in nutrient composition in DDGS could be considered as possible solutions for improving pork fat firmness.

Since the oil and linoleic acid content of DDGS is the primary reason for reduced pork fat firmness, it is reasonable to assume that the increase in oil extraction from DDGS would improve pork fat firmness. However, it appears that reducing DDGS in the diet (from 40 to 20%) has a greater effect on improving jowl, backfat, and belly fat firmness (as observed by decreasing iodine value) than reducing the oil content of DDGS (from 9.6 to 5.4% oil DDGS; Graham et al., 2014). This could be attributed to changes in ADFI because when reducing the DDGS oil content, pigs had greater ADFI which presumably resulted in similar lipid and linoleic acid intake. These results are also contrary to the assumption that fiber in DDGS is a limiting factor for feed intake. It may be possible that the high lipid content in DDGS decreases ADFI, or perhaps there is an interactive effect between the lipid and fiber content of DDGS. As mentioned previously, fiber, and the interaction between fiber and lipid, are poorly understood in the pig. The observation by Graham et al. (2014) also agrees with the findings by Wu et al. (2014), who observed a decrease in ADFI when feeding diets with 40% DDGS of 9.9 and 14.2% oil, and similar ADFI when feeding diets with 40% DDGS containing 5.9% oil compared to pigs fed corn-soybean meal diets. In that study, iodine value of backfat, belly fat, and jowl fat was unchanged when reducing the DDGS oil content from 5.9% and 9.9% oil. However, when the DDGS oil content was 14.2%, all depots had greater iodine value compared to pigs fed DDGS with 5.9% and 9.9% oil (unpublished data). Both of these studies indicate that reducing the oil content of DDGS from about 10% to about 5.5% has a limited effect on improving pork fat firmness and supports that concept that overall

lipid and linoleic acid intake is important to consider. The importance of focusing on linoleic acid intake has also been recently proposed in predicting fatty acid composition of pork carcass fat by Kellner (2014).

Withdrawing DDGS from the diet for a minimum of 3 weeks before harvest has been effective at improving fat firmness of belly fat (as indicated by reduced carcass fat iodine value), but belly fat IV was not fully restored to the IV observed in pigs fed corn-soybean meal diets until a 9 week withdrawal period was used (Xu et al., 2010a). Similarly, withdrawing DDGS for 20 or 40 days, or for the entire growing-finishing period, resulted in a linear decrease of backfat, belly fat, and jowl fat IV compared to pigs fed 30% DDGS diets throughout the entire growing-finishing period (Jacela et al., 2009). This response did not occur when dietary DDGS inclusion rate was reduced from 30 to 15% for 20 or 40 days before harvest (Jacela et al., 2009). Other commercial approaches to control fatty acid intake of pigs during the growing-finishing period have included the use of a "step-down" or gradual decrease in dietary DDGS inclusion rates, but the effectiveness of using this strategy has not been evaluated experimentally. Several have used dietary fatty acid composition and length of feeding dietary fat sourcesto predict carcass fatty acid composition, and use these predictions a constraint when formulating swine diets.

2. Fatty acid composition prediction

Several carcass iodine value prediction equations have been developed using linoleic acid intake (Averette Gatlin et al., 2002; Kellner, 2014), dietary linoleic acid concentration (Benz et al., 2011a), dietary DDGS inclusion rate (Cromwell et al., 2011), daily iodine value of the product, calculated as (percentage of dietary lipid) × (iodine

value of dietary lipid) × 0.1; **IVP**; Madsen et al., 1992) intake (Madsen et al., 1992; Kellner, 2014), dietary IVP (Benz et al., 2011a; Kellner, 2014) to estimate subsequent IV for different pork fat depots (Table 1.10).

Iodine value product was first proposed in 1962 to calculate not only lipid content, but also specific fatty acid composition of feed ingredients. Much like iodine value, IVP is a composite value of fatty acid composition and includes the amount of dietary lipid present in the fat source. Benz et al (2011a) formulated diets based on IVP in an attempt to predict carcass fat depot iodine values, and concluded the use of IVP was a poor predictor of backfat and jowl fat IV when choice white grease (saturated fatty acid source) was included in diets formulated to achieve a "high IVP". As a result, the authors concluded that calculated dietary linoleic acid content was a better predictor of carcass fat IV in both jowl fat and backfat (Table 1.10; Benz et al., 2011a). Testroet et al., 2014) indicated that, "jowl fat iodine values are significant predictors of back and belly fat iodine values", but the reported $R^2 \leq 0.36$ suggests they are not reliable predictors. Benz et al. (2010) stated that "jowl fat iodine value can determine the overall trend in changes in iodine value", however, changes in iodine value are not equal across fat depots as previously discussed. While some packers use the jowl location to determine iodine value, the research results do not support that fatty acid profiles of jowl fat are consistent with fatty acid profiles in belly fat. The addition of 30% DDGS to swine diets usually doubles the linoleic acid content of pork fat (Asmus et al., 2014b, Cromwell et al., 2011, Xu et al., 2010b, Xu et al., 2010a). Since linoleic acid intake has such a profound effect on fatty acid composition of pork fat, is seems logical that linoleic acid intake should be a primary focus for controlling fatty acid composition of pork fat.

Table 1.10. Summary of regression equations developed to predict jowl, back, or belly fat carcass iodine value

Study	Jowl	Back	Belly	Equation	*P* value	R^2
Averette Gatlin et al., 2002		X[1]		Y = 1.76 + 0.031 (linoleic acid intake/d)	NA	0.70
Cromwell et al., 2011		X		Y = 0.432x + 64.5, where x = % of DDGS in the diet from 0 to 45%	NA	0.917
Kellner, 2014	Average across 3 depots			60.58 + [0.121 × 18:2n-6 intake/d (g)]	< 0.01	0.611
Kellner, 2014	Average across 3 depots			62.55 + [3040.4 × 18:3n-3 intake/d (g)]	< 0.01	0.26
Kellner, 2014	Average across 3 depots			72.94 - [0.06 × 16:0 intake/d (g)]	0.03	0.08
Kellner, 2014	Average across 3 depots			72.13 - [0.135 × 18:0 intake/d (g)]	0.01	0.12
Kellner, 2014	Average across 3 depots			66.30 - [0.085 × 18:1 intake/d (g)]	0.02	0.08
Kellner, 2014	Average across 3 depots[2]			70.34 + [0.002 × IVP intake/d (g)]	0.83	< 0.01
Paulk et al., 2014	X[3]			85.50 + (1.08 × I EFA) + (0.87 × F EFA) - (0.014 × I d) - (0.050 × F d) + (0.038 × I EFA × I d) + (0.054 × F EFA × F d) - (0.0066 × I NE) + (0.071× I BW) - (2.19 × ADFI) - (0.29 × backfat depth)		
Paulk et al., 2014		X[3]		84.83 + (6.87 × I EFA) - (3.90 × F EFA) - (0.12 × I d) - (1.30 × F d) - (0.11 × I EFA × F d) + (0.048 × F EFA × I d) + (0.12 × F EFA × F d) - (0.0060 × F NE) + (0.0005 × F NE × F d) - (0.26 × backfat depth)		
Paulk et al., 2014			X[3]	106.16 + (6.21 × I EFA) - (1.50 × F d) - (0.11 × I EFA × F d) - (0.012 × I NE) + (0.00069 × I NE × F d) - (0.18 × HCW) - (0.25 × backfat depth)		
Madsen et al., 1992		X[2]		47.1 + 0.14 × IVP intake/d; where dietary IVP ranged from 37 to 88	NA	0.86
Benz et al., 2011a	X[2]			0.247 × Dietary IVP + 56.479; where IVP ranged from 37.1 to 55.3	0.24	0.321
Benz et al., 2011a		X[2]		0.2715 × Diet ary IVP + 51.946; where IVP ranged from 37.1 to 55.3	0.44	0.157
Benz et al., 2011a	X			10.111 × Dietary 18:2n6 + 47.469; where dietary linoleic acid ranged from 1.70 to 2.47%	< 0.01	0.903

Benz et al., 2011a	X	14.324 × Dietary 18:2n6 + 35.458 ;where dietary linoleic acid ranged from 1.70 to 2.47%	< 0.03	0.734

[1] Backfat linoleic acid content
[2] Iodine value of the product (IVP)
[3] I = initial diet, F = final diet, d = duration of diet fed, EFA = essential fatty acids, NE = net energy, HCW = hot carcass weight

Key pieces of information are lacking in implementing iodine value as a market hog procurement specification. Incremental changes in pork fat quality must be related to economic losses due to poor fat quality. Establishing this economic relationship will help identify appropriate methods for fat quality quantification and establish pork production targets. If producers do not have pork fat quality targets to achieve, production decisions by producers and diet formulation by nutritionists will remain focused on lowest cost of production. Alternatively, the negative effects of feeding DDGS on carcass dressing percentage and pork fat firmness can be accomplished by specific feeding strategies.

IX. Effect of feeding DDGS to immunologically castrated pigs on pork fat quality and carcass dressing percentage

The general, the decline in lipogenesis and greater deposition of fatty acids from dietary fat sources is of significant importance in immunologically castrated pigs because they have less backfat compared to physically castrated pigs (Table 1.3) and would likely be more affected by dietary fatty acid changes (Wood et al., 1989). Additionally, linoleic acid is the primary contributor to soft pork fat in pigs fed DDGS diets because of its high degree of unsaturation and abundance in DDGS (NRC, 2012). Linoleic acid, and other polyunsaturated fatty acids, are very susceptible to peroxidation and the production of off-flavors and odors in meat products (Wood et al., 2008). Since immunologically castrated pigs can be harvested 3 to 10 weeks after the second dose of Improvest®, fat deposition increases during this time period. Theoretically, pigs marketed soon after the second dose of Improvest® would be most sensitive to dietary changes in fatty acid intake and would benefit from alternative DDGS feeding strategies, such as withdrawing or gradually decreasing dietary DDGS content before harvest.

Only 1 study has been conducted to evaluate feeding diets containing 30% DDGS to immunologically castrated pigs (Asmus et al., 2014b). Feeding diets containing 30% DDGS resulted in lower dressing percentages in immunologically castrated pigs compared to physically castrated pigs (Asmus et al., 2014b). Withdrawing DDGS for 5 or 7 weeks before harvest restored dressing percentage of immunologically castrated pigs to levels similar as pigs fed corn-soybean meal diets (Asmus et al., 2014b). By increasing the time interval between the second Improvest® dose and harvest from 5 to 7 weeks, backfat and belly fat iodine value decreased more dramatically in immunologically castrated pigs than in physically castrated pigs (Asmus et al., 2014b). However, iodine value of jowl fat from immunologically castrated pigs did not change by increasing the interval between the second dose and harvest from 5 to 7 weeks (Asmus et al., 2014b). These results suggest that in immunologically castrated pigs, jowl fat is less responsive to changes in dietary fatty acids than backfat and belly fat, and that use of a withdrawal feeding strategy is effective in lowering carcass fat iodine value. However, in order to achieve a belly fat iodine value similar to pigs fed corn-soybean meal diets, the marketing window between the second Improvest® dose and harvest needs to be increased to 7 weeks.

X. Current unknowns of feeding DDGS to immunologically castrated pigs

Only 1 study has been published on the effects of feeding DDGS diets to immunologically castrated pigs, and focused on a DDGS withdrawal feeding strategy following the feeding of 30% DDGS and harvesting 5 or 7 weeks after the second Improvest® dose (Asmus et al., 2014b). Harvesting pigs at 5 or 7 weeks after the second Improvest® dose only represents a small portion of the 3 to 10 week time period after the

second dose of Improvest® that pigs can be harvested. The withdrawal feeding strategy used by Asmus et al. (2014) was effective at improving the iodine value of belly and backfat. However, at times the price relationship between corn, soybean meal, and DDGS favors DDGS use exceeding 30% in growing-finishing pig diets. Feeding diets containing greater than 30% DDGS to immunologically castrated pigs has not been evaluated, nor has the use of common DDGS feeding strategies (such as gradually decreasing dietary DDGS levels). In addition, the use of different feeding strategies may alter the optimum time to market pigs within the 3 to 10 week time period after the second Improvest® dose.

In summary, the use of Improvest® can achieve several sustainability goals of pork production through improving resource and environmental sustainability as a result of reducing feed intake, improving energy and nutrient utilization efficiency, reducing carcass fat, and reducing manure nitrogen excretion. However, the use of Improvest® in conjunction with other cost effective pork production methods, such as high dietary inclusion levels of DDGS and use of DDGS feeding strategies, needs to be evaluated to optimize the growth performance benefits observed in immunologically castrated pigs as well as the pork quality needs of pork packers and processors. Complementary DDGS feeding strategies and Improvest® management strategies need to be identified that balance the needs of pork producers, packers, and processors. This will also result in the production of more high quality pork with fewer resources and promote economic and environmental sustainability while feeding more consumers.

XI. Statement of the problem

The increasing global population of consumers will require greater amounts and more efficient lean meat production. The use of temporary immunological castration in pigs allows for pigs to grow as intact male pigs for a longer period of time than physical castrates, while reducing boar taint in pork products. Immunologically castrated pigs have less overall feed intake, less adipose accretion, improved energy and nutrient utilization, and produce lean pork more efficiently. A reduction in the time interval between the second Improvest® dose and harvest results in pigs with less carcass fat. However, pigs with less carcass fat have a higher concentration of polyunsaturated fatty acids in fat depots, resulting in undesirable soft pork fat. Soft pork fat in pork products can create handling, processing, and shelf-life challenges. Use of DDGS in swine growing-finishing diets has several advantages, but the primary limitation is its effect on reducing pork fat firmness due to the high linoleic acid content of corn oil of DDGS. In fresh pork products, the belly and loin primals are of greatest concern due to their high value, particularly for the belly because of its relatively high fat content and need for further processing into bacon.

In order to apply the use of immunological castration to modern pork production systems where DDGS is widely used, feeding strategies need to be identified, along with finding the optimum interval between second Improvest® dose and harvest, to overcome the possible negative interactive effects of feeding DDGS to immunologically castrated pigs. In order to achieve this objective, producer, packer, and processor needs must be considered relative to impacts on growth performance, changes in body and carcass composition, and lean and fat quality of pork carcasses. The beneficial characteristics of

immunological castration and use of DDGS in diets during the growing-finishing period may provide an economic, environmental, and social benefit to sustaining the production and consumption of lean pork for both producers and consumers.

CHAPTER 2: Growth performance of immunologically castrated pigs harvested at 5, 7, or 9 weeks after the second Improvest® dose and fed diets containing corn dried distillers grains with solubles

I. Summary

Growth performance of immunologically castrated (**IC**) pigs (n = 863) was determined when increasing time intervals between the second Improvest® (*gonadotropin releasing factor (**GnRF**) analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest (**TD**), and with different DDGS feeding strategies (**FS**) in a 4 × 3 factorial arrangement of treatments. Pigs were fed 1 of 4 dietary feeding strategies for 16 wk and included: 1) corn-soybean meal control diets (**PCon**), 2) a gradual decrease of dietary DDGS inclusion (**SD**) rate from 40, 30, 20, and 10% in phases 1 to 4, respectively, 3) feeding 40% DDGS diets in phases 1 to 3, and removal of DDGS from the phase 4 diet (**WD**), and 4) feeding 40% DDGS diets in all 4 phases (**NCon**). Pigs received the second Improvest® dose at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest. There were no interactions between FS × TD × wk for any measure of growth performance. Pigs fed PCon and WD had greater ($P < 0.05$) overall ADFI than pigs fed NCon, especially when harvested 9 wk after the second Improvest® dose (2.45 and 2.44 vs. 2.31 ± 0.08 kg/d, respectively). This response was partly due to withdrawing DDGS from the diet at 19 wk of age for pigs fed the WD feeding strategy, which led to a tendency ($P < 0.10$) for pigs fed WD to have increased ADFI between the 19 to 21 wk and 21 to 24 wk intervals (3.26 vs. 3.51 ± 0.09 kg/d, respectively). During the same time period, ADFI was unchanged ($P > 0.05$) in pigs fed PCon, SD, and NCon. Overall G:F was improved ($P < 0.05$) in TD5 pigs compared with TD9 pigs, and tended ($P < 0.10$) to be improved compared with TD7

pigs (0.428 vs. 0.413 and 0.417 ± 0.003, respectively). These changes in growth performance resulted in diverging BW of pigs beginning in phase 3, where pigs fed PCon had greater ($P < 0.05$) BW than pigs fed WD and NCon. At the beginning of phase 4, pigs fed PCon had greater ($P < 0.05$) BW than all other feeding strategies. Final BW was similar among pigs fed SD, WD, and PCon. Throughout the growing-finishing period, BW was similar among TD treatments. The SD feeding strategy was more effective than the WD feeding strategy in maintaining overall G:F (0.424 and 0.414 ± 0.005, respectively) and ADG (0.94 and 0.93 ± 0.03 kg/d, respectively) similar ($P > 0.05$) to pigs fed PCon (0.427 ± 0.005 and 0.96 ± 0.03 kg/d, respectively). Differences in final BW among feeding strategies, and lack of BW differences among TD treatments were explained by differences in body composition (Chapter 4).

KEYWORDS: carcass characteristics, dried distillers grains with solubles, feed intake, feeding strategy, immunological castration, pigs

II. Introduction

Intact male (**IM**) pigs have lower feed intake, greater lean gain efficiency, and lower cost of lean gain compared with physical castrates (**PC**; Squires, 2011). Historically in the U.S., IM pigs have been physically castrated at an early age to avoid unpalatable off-odors, known as boar taint (Sutherland et al., 2010). Immunological castration (**IC**) captures the improved lean gain efficiency of IM pigs while minimizing boar taint. The second Improvest® (*gonadotropin releasing factor (**GnRF**) analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose is administered at least 4 wk after the first dose and 3 to 10 wk before harvest (FDA, 2011a). Increasing the

interval between the second Improvest® dose and harvest results in a linear increase in overall ADFI (Lealiifano et al., 2011).

Feeding corn dried distillers grains with solubles (**DDGS**) can decrease overall ADFI when increasing dietary levels of DDGS up to 45% (Cromwell et al., 2011), and 60% (Hardman, 2014; Bergstrom et al., 2009a). Additionally, feeding DDGS can reduce carcass dressing percentage and result in undesirable, soft pork fat in PC and gilts which creates processing challenges (Stein and Shurson, 2009). Reductions in carcass dressing percentage and pork fat firmness can be overcome by withdrawing DDGS from the diet before harvest (Gaines et al., 2007; Xu et al., 2010a). Only 1 study has evaluated the use of a withdrawal feeding strategy (**FS**) in IC pigs fed diets containing 30% DDGS (Asmus et al., 2014b). The dietary inclusion of DDGS can exceed 30% to reduce feed cost when prices are favorable relative to corn and soybean meal. However the effectiveness of withdrawing or gradually reducing the DDGS have not been evaluated in IC pigs harvested within the 3 to 10 wk time period after the second Improvest® dose. Therefore, the objectives of this study were to determine feed intake and growth performance patterns of IC pigs harvested at 5, 7, or 9 wk after the second Improvest® dose when using different DDGS FS.

III. Materials and methods

All animal care and use procedures were approved by the University of Minnesota Institutional Animal Care and Use Committee (#1112B07511).

A. Animals and housing

This study was conducted at the University of Minnesota West Central Research and Outreach Center (Morris, MN). Four groups of IM pigs (n = 863; V100 Landrace

females x V40 Large White boars; Genetiporc, Alexandria, MN) were weighed (initial BW = 21.5 ± 0.8 kg) and individually identified using ear tags at 8 weeks of age (**WOA**). Pigs were allotted randomly to one of 12 treatments in a 4 x 3 factorial arrangement, with 4 feeding strategy treatments and 3 Improvest® treatments (Figure 2.1). Pigs were housed in an environmentally controlled growing-finishing barn containing 24 pens with fully slatted floors, one nipple waterer, and one 4-hole feeder. Each pen (4.66 x 1.58 m) housed 9 pigs. All pig morbidity, type and duration of medications provided, and mortality were recorded. The 4 groups of pigs were harvested in July and September of 2012, and January and May 2013. All pigs (n = 835) were harvested at 24 WOA. A subsample of pigs (n = 192) were harvested at the University of Minnesota Meat Science Laboratory and the remainder of pigs (n = 643) were harvested at a commercial abattoir.

B. DDGS feeding strategies

Four dietary feeding strategies were used in a 4-phase feeding program during the 16 wk growing-finishing period. Phase 1 diets were fed for 3 wk, phase 2 and 3 diets were each fed for 4 wk, and phase 4 diets were fed for 5 wk. Dietary phase changes occurred at a mean BW of 35.8, 60.1, and 88.4 kg in phases 2, 3, and 4, respectively. Feeding strategies included: positive control (**PCon**) where pigs were fed 0% DDGS, corn-soybean meal based diets; DDGS step down (**SD**) where pigs were fed 40%, 30%, 20%, and 10% DDGS diets in the 4 dietary phases, respectively; DDGS withdrawal (**WD**) where pigs were fed 40% DDGS diets in phases 1 to 3, and a 0% DDGS diet in phase 4; and negative control (**NCon**) where pigs were fed 40% DDGS diets throughout the entire growing-finishing period (Tables 2.1 and 2.2). All pigs had ad libitum access to feed and water.

C. Immunological castration treatments

All pigs were immunologically castrated by receiving two, 2-mL doses of Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) via s.c. injections at the post-auricular region of the neck by Zoetis, Inc. trained technicians. The first dose of Improvest® was administered to all pigs at 11 WOA. The second dose of Improvest® was administered at 15, 17, or 19 WOA, which corresponded with 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest, respectively. Two wk following the second Improvest® dose, a quality assurance assessment was performed according to the Zoetis, Inc protocol to assure successful immunological castration. Each pig was individually evaluated for "boar-like" behavior, as well as testes size and scrotum color (FSIS, 2013). Pigs failing to meet these specifications (n = 38) received a third dose of Improvest® and remained in the study.

D. Diet formulation

A mathematical model was used for estimating nutrient requirements for lean growth IM pigs (NRC, 2012). Feed intake for each dietary phase was calculated using previous feed intake data of PC and gilts fed in the same environment at the University of Minnesota West Central Research and Outreach Center, which was then adjusted using published relative feed intake differences between IC and PC pigs (Dritz et al., 2011). Nutrient requirements for standardized ileal digestible (**SID**) Lys, Ca, and P were increased by 5% to include a safety margin to assure IM pigs would express their full growth potential. Diets were not isocaloric because achieving this would have required the addition of a supplemental fat source and thus, supplemental fat would be a confounding factor when determining changes in pork fat quality among treatments. All

diets were formulated to contain similar SID Lys to ME ratios and Ca:P among phases (Table 2.1 and 2.2), and diets were fed in meal form. Since the P present in DDGS is more digestible than in corn and soybean meal, two basemixes (one for 0% DDGS diets and one for 40% DDGS diets) were used to supply pigs with required Ca, P, and other vitamins and minerals. The two basemixes were blended proportionately when diets contained 30, 20, or 10% DDGS.

NRC (2012) nutrient composition tables were used to determine ME, SID AA, and available P, along with other nutrients for corn and soybean meal (Table 2.3). Estimates of ME, SID AA, and available P content of this DDGS source were obtained from Illuminate® (Value Added Science and Technology, Mason City IA) for use in diet formulation (Table 2.3). A single source of dried distillers grains with solubles was obtained and contained 10.4% ether extract (Table 2.4 and 2.5).

Two wk before harvest, group 1 pigs were diagnosed with Hemorrhagic Bowel Syndrome (**HBS**), which was confirmed by necropsy. This prompted the need to orally administer lincomycin (Zoetis, Inc. Florham Park, NJ) until harvest. Group 1 was administered lincomycin through the water for 2 wk before harvest. Group 2 pigs were housed concurrently and were also orally administered lincomycin for 3 wk through the water beginning at 16 WOA. Once the dietary phase changed from phase 3 to 4, diets included 0.044 g/kg diet of lincomycin for 5 wk. Due to the known health history of this source of pigs, group 3 pigs were administered 0.044 g/kg diet of lincomycin throughout the entire growing-finishing period. However, 10 d before harvest of group 3 pigs, another mortality occurred due to HBS, resulting in an increased dosage of feed administered lincomycin to 0.088 g/kg diet. Group 4 pigs were fed 0.088g/kg diet of

lincomycin throughout the growing-finishing period with the addition of water-medicated Denagard® for 6 d at 10 WOA to treat Ileitis.

E. Dietary composition

One mixing lot of each experimental diet for each phase and group of pigs was subsampled and analyzed for nutrient content. Reported dietary composition values include the mean of four mixing lots for each diet. Proximate analysis, AA (Table 2.5), and fatty acid (Table 2.4) profiles of the DDGS source used, as well as the experimental diets (Tables 2.6 and 2.7) were determined by the University of Missouri Agricultural Experiment Station Chemical Laboratories (Columbia, MO) using AOAC method 934.01 for moisture, AOAC method 920.39 (A) for ether extract, AOAC method 942.05 for ash, AOAC method 984.13 (A-D) for crude protein, AOAC method 973.18 (A-D) for ADF, JAOAC 56, 1352-1356, 1973 for NDF, AOAC method 982.30 E (a-c) for AA profile, and AOAC methods 965.49 and 996.06 for fatty acid profile analysis. Dietary iodine value product (**IVP**) was calculated from the analyzed lipid content and fatty acid composition of each diet using the formula: IVP = IV of dietary fat × percentage of dietary fat × 0.10 (Madsen et al., 1992), where the iodine value of dietary fat was calculated from fatty acid analysis ([C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723; AOCS, 1998).

F. Growth performance

Pigs were weighed individually at the beginning and end of each dietary phase and 2 wk after the beginning of phases 2 to 4 to determine weight gain for each pen. Feed offered during each period was recorded, and at the end of each weigh period, the remaining feed was removed, weighed, and subtracted from the total feed offered to

determine total feed disappearance on a pen basis for each weigh period. From these data, ADFI, ADG, and gain efficiency (G:F) were calculated.

G. Statistical analysis

Growth performance was analyzed using PROC MIXED in SAS (Cary, NC) where pen was considered the experimental unit. The statistical model included diet, TD, and diet × TD as fixed effects, and group as a random effect. Week was included as a repeated measure in the data analysis. The residuals were used to test the assumptions of normality using PROC Univariate. When the Kolmogorov-Smirnov test for normality was significant ($P < 0.05$), any datapoint with a residual > ± 4, was removed from analysis. This included 3 datapoints for ADFI and 2 datapoints for G:F. Least squares means were separated and adjusted using the Tukey option. Significance was declared when $P \leq 0.05$ and trends were identified when $P \geq 0.05$ and ≤ 0.10.

IV. Results and discussion

A. Pig health

Four pigs were removed from the study. One pig was determined to be a cryptorchid after the beginning of the study, 1 pig was removed due to severe weight loss and diarrhea, 1 pig was removed due to low weight gains in multiple weigh periods, and 1 pig was removed due to navel sucking.

B. Ingredient and dietary composition

Nutrient composition estimates obtained from Illuminate® for the DDGS source used in diet formulation were slightly lower than the analyzed nutrient content of the same DDGS that was fed (Table 2.5). Since nutrient composition can vary among and

within DDGS sources, we attempted to minimize variation in nutrient content by using a single source of DDGS. The ether extract (crude fat) content of this DDGS source was representative of traditionally produced, high oil-DDGS (NRC, 2012). While the majority of ethanol plants are extracting corn oil prior to producing DDGS, we chose to use a high-oil DDGS source because it contains a high concentration of PUFA, which compared with feeding corn-soybean meal diets, reduces pork fat firmness when fed at high (> 20%) dietary inclusion rates to growing-finishing PC and gilts (Xu et al., 2010b). The analyzed ether extract content of the DDGS source was 10.4% and the ether extract contained 53.9% linoleic acid (Tables 2.5 and 2.4). The NDF and ADF content of the DDGS source used was 34.10, and 12.30%, respectively (Table 2.5). The high concentration of linoleic acid in corn oil present in DDGS, directly affects the fatty acid composition of pork adipose depots. One of the overall objectives of conducting this study was to determine the effectiveness of withdrawing DDGS or gradually decreasing the DDGS dietary inclusion rate during the growing-finishing period on pork fat quality. In this study, we wanted DDGS to be the primary contributor of dietary lipid. Therefore, formulated diets were not isocaloric because this could have only been achieved by using a source of supplemental fat, which would have confounded the pork fat quality responses. Due to the inherent compositional differences between experimental diets used in this study, they were formulated to have a similar SID Lys:ME to ensure adequate SID AA intake (Tables 2.1 and 2.2).

C. Feed intake

Feed intake of IC pigs increases rapidly beginning at d 5 to 6 after the second Improvest® dose, so that by d 13 to 14 after the second Improvest® dose, ADFI of IC

pigs exceeds that of PC pigs by 7% (Elsbernd et al., 2014). Average daily feed intake continually increases, and by 21 and 28 d after the second Improvest® dose, ADFI has been reported to be 10 and 15% greater than PC pigs, respectively (Elsbernd et al., 2014). In the present study, there were no 3-way interactions between feeding strategy, timing of the second Improvest® dose, or wk ($P > 0.05$) for any measure of growth performance. Even though there were no 3-way interactions, marginal ADFI differences throughout the growing-finishing period accumulated and led to a tendency ($P < 0.10$) for reduced overall ADFI of TD9 pigs fed NCon compared with TD9 pigs fed PCon and WD (2.31 vs. 2.45 and 2.44 ± 0.08 kg, respectively; Figure 2.2 and Appendix Table A.1). As a result, there was a tendency for pigs fed NCon to have reduced ($P < 0.10$) overall ADFI compared with pigs fed PCon (2.40 vs. 2.34 ± 0.07 kg, respectively; Table 2.8).

As the finishing period progressed, pigs with longer time intervals between the second Improvest® dose and harvest appeared to have an earlier peak for ADFI than those with shorter time intervals (Figure 2.3 and Appendix Table A.3). In TD9 pigs, ADFI, peaked during the 17 to 19 wk interval and was not different between 17 to 24 WOA. In TD7 pigs, ADFI peaked during the 19 to 21 wk interval and was not different between 19 to 24 WOA. In TD5 pigs, ADFI continually increased throughout the finishing period. These differences resulted in TD5 pigs having lower ($P < 0.05$) ADFI during the 17 to 19 wk interval compared with TD9 pigs, while ADFI of TD7 pigs was intermediate ($P > 0.05$) to both TD9 and TD5 pigs (2.64 vs. 2.95 and 2.48 ± 0.10 kg, respectively) during the 17 to 19 wk interval. Within time interval, these differences were expected because TD9 pigs received the second Improvest® dose at 15 WOA and were IM pigs for a shorter period of time. During the 19 to 21 wk interval, TD5 pigs had lower

($P < 0.05$) ADFI compared with TD9 and TD7 pigs (2.84 vs. 3.25 and 3.37 ± 0.10 kg, respectively). Since the second Improvest® dose was administered to TD5 pigs at 19 WOA, these pigs were IM pigs for a longer period of time compared with TD7 and TD9 pigs. Therefore, lower ADFI was expected in TD5 pigs compared to TD7 and TD9 pigs. The rapid increase ($P < 0.05$) of ADFI of TD5 pigs during the 21 to 24 wk interval resulted in all TD treatments having similar ($P < 0.05$) ADFI during the final 3 wk period. Changes in feed intake of IC pigs over time have only been evaluated by Lealiifano et al. (2011). In that study, the rate of ADFI increased in pigs receiving the second Improvest® dose 6 wk before harvest slowed, and was not different 3 to 6 wk after the second dose. The time interval between the second dose and harvest evaluated by (Lealiifano et al., 2011) were too short (≤ 4 wk) to observe a plateau in ADFI, but in the current study, ADFI plateaued in TD treatments that were ≥ 7 wk.

The hormonal shift that occurs following the second Improvest® dose is associated with regulation of feed intake. Intact male pigs produce large amounts of testosterone and estradiol (Clapper et al., 2000), both of which reduce feed intake (Claus and Weiler, 1994). Within 5 to 10-d after the second Improvest® dose, plasma LH and testosterone rapidly decrease (Claus et al., 2007). This corresponds with the timing of the initial rise in ADFI reported by Elsbernd et al. (2014). In this study, the increase in ADFI of TD9 and TD7 pigs after the second Improvest® dose was similar to pigs that had not yet received the second Improvest® dose. For example, TD9 pigs which received the second Improvest® dose at 15 WOA had similar ADFI compared with TD7 and TD5 pigs during the 15 to 17 wk interval (2.44 vs. 2.35 and 2.36 ± 0.10 kg, respectively; Figure 2.3 and Appendix Table A.2). Additionally, TD7 pigs which received the second

dose at 17 WOA had similar ADFI compared with TD5 pigs during the 17 to 19 wk interval (2.64 vs. 2.48 ± 0.10 kg, respectively). Intact male and IC pigs have greater visceral mass than PC pigs (Boler et al., 2014) presumably due to androgen-stimulated visceral growth before the second Improvest® dose is administered. Given the younger age of TD9 pigs, compared with TD7 and TD5 pigs, when they received the second Improvest® dose, TD9 pigs could have had more limited gut capacity when receiving the second Improvest® dose than TD7 or TD5 pigs. This could have limited the expected rapid increase in ADFI associated with the hormonal shift following immunological castration. Thus, TD9 pigs may have been particularly sensitive to high fiber diets containing 40% DDGS because the presumed visceral growth that occurs in IC pigs may have been limited. However, Asmus et al. (2014) fed diets containing corn-soybean meal, corn-soybean meal diets containing 30% DDGS, or corn-soybean meal diets where 30% DDGS was removed for 5 or 7 wk before harvest, and observed no difference in overall ADFI of IC pigs. Data from this study along with data from Lealiifano et al. (2011) suggests that the magnitude of ADFI increase may be related to age when the second Improvest® dose is administered, length of time between the second Improvest® and harvest, and dietary fiber concentration.

The interactive effect of overall feed intake (Table 2.8 and Appendix Table A.1) and the lack of a TD main effect (Table 2.8) could be linked to the high fiber, high CP content of DDGS diets fed in this study. Feeding the WD strategy tended ($P < 0.10$) to increase ADFI between the 19 to 21wk and 21 to 24 wk intervals (3.26 vs. 3.51 ± 0.09 kg, respectively; Figure 2.3 and Appendix, Table A.3), so that during the 19 to 21 wk interval, pigs fed WD had greater ($P < 0.05$) ADFI compared with pigs fed SD and NCon

(3.26 vs. 3.10 and 3.11 ± 0.09 kg, respectively). Pigs fed PCon had similar ADFI compared with all feeding strategies during the 19 to 21 wk interval. During the 21 to 24 wk interval, pigs fed WD had greater ($P < 0.05$) ADFI compared with all other feeding strategies (3.51 vs. 3.35, 3.32, and 3.30 ± 0.09 kg, respectively). This increase in ADFI (Figure 2.3 and Appendix Table A.3) occurred when pigs switched from consuming a diet containing 3,232 kcal/kg to 3,333 kcal/kg (Table 2.2). These results are contrary to the common assumption that pigs will voluntarily consume feed to achieve a similar energy intake, regardless of the dietary energy density (Ellis and Augsperger, 2001). As a result, there are likely other factors that control feed intake when feeding diets containing 40% DDGS, and withdrawing 40% DDGS 5 wk before harvest. There are many dietary and physiological factors that influence voluntary feed intake (Black et al., 2009). It is possible that feed intake of pigs fed NCon and WD was limited by the greater fiber content of the diets containing 40% DDGS. Feeding high fiber diets can result in greater intestinal fill due to the bulk density and water binding effect of fiber (Kerr and Shurson, 2013). Abruptly reducing the DDGS inclusion from 40 to 0% would allow for greater capacity of feed intake if the diet has a lower bulk density. Similarly, Hilbrands et al. (2013) reported lower ADFI when feeding diets containing 40% DDGS compared with feeding diets containing 20% DDGS. Abruptly switching pigs from diets containing 40% DDGS to a corn-soybean meal diet restored ADFI to levels similar to pigs fed corn-soybean meal or 20% DDGS (Hilbrands et al., 2013).

D. Average daily gain and body weight

In the present study, overall ADG decreased ($P < 0.05$) in pigs fed NCon compared with pigs fed PCon and tended to decrease ($P < 0.10$) compared with pigs fed

SD (Table 2.8). The decrease in overall ADG of pigs fed NCon observed in this study began during the 13 to 15 wk interval when ADG was lower ($P < 0.05$) compared with pigs fed PCon (Figure 2.4 and Appendix Table A.4). Similar to the current study, Cromwell et al. (2011) fed diets containing up to 45% DDGS to PC and gilts and observed a linear decrease in ADG during phase 1 and the overall (approximately 13 wk) feeding period. Hardman et al. (2014) observed a linear decrease in overall ADG when feeding diets containing up to 60% DDGS. Additionally, feeding diets with 60% DDGS decreased ADG compared with feeding diets with 20% DDGS (Bergstrom et al., 2009b). However, feeding diets to pigs with 30 or 60% DDGS did not alter ADG (Weber et al., 2013). It should be noted that corn-soybean meal control diet were not included in the Weber et al. (2013) study. However, pigs fed 30 and 60% DDGS diets (Weber et al., 2013) had similar overall ADG to pigs fed 30 and 45% DDGS reported by Cromwell et al. (2011), and similar overall ADG to pigs fed 40% DDGS in the present study. The variable response of ADG when pigs are fed less than 30% DDGS is presumably due to the variation of DDGS nutrient composition and the use of generic compositional estimates for diet formulations (Stein and Shurson, 2009). As a result, increasing the diet DDGS inclusion rate above 30% would only magnify this problem. Hilbrands et al. (2013) demonstrated that reductions ADG of pigs fed 40% DDGS were more apparent in PC and gilts when feeding DDGS sources with low SID AA. However, feeding 40% DDGS with high SID AA resulted in similar ADG to pigs fed corn-soybean meal diets (Hilbrands et al., 2013). Although few studies have evaluated feeding DDGS in excess of 30% DDGS of the diet to growing-finishing pigs, it appears feeding more than 30%

DDGS consistently results in reduced ADG compared with pigs fed corn-soybean meal, especially when feeding DDGS sources with low SID AA.

The changes in ADG observed in the present study resulted in BW of pigs deviating at the beginning of phase 3 where pigs fed NCon and WD had reduced ($P < 0.05$) BW compared with pigs fed PCon (59.3 and 59.0 vs. 62.2 ± 0.9 kg, respectively; Table 2.9). At the beginning of phase 4, pigs fed PCon had greater ($P < 0.05$) BW compared with pigs fed SD, WD, and NCon (91.7 vs. 88.6, 86.8, and 86.4 ± 0.9 kg, respectively). Final live weight of pigs fed PCon was greater ($P < 0.05$) than pigs fed WD and NCon (125.3 vs. 122.3 and 120.0 ± 0.09 kg, respectively). Similarly, Cromwell et al. (2011) observed a linear decrease in BW when feeding diets containing up to 45 % DDGS to gilts and PC from approximately 60 to 118.5 kg. Likewise, Hardman et al. (2014) observed a linear decrease in BW beginning at approximately 33.8 kg when feeding up to 60% DDGS diets. The reduction in ADG and BW observed in NCon pigs relative to other feeding strategies is likely due to the observed reduction in feed intake. This could have also reduced energy and nutrient intake which is discussed in Chapter 3.

The removal of DDGS from the diet at 19 WOA (WD) resulted in greater ($P < 0.05$) ADG compared with pigs fed PCon (1.18 vs. 1.09 ± 0.03 kg, respectively), but this only occurred in the first 2 wk after DDGS withdrawal (Figure 2.4 and Appendix Table A.4). This result is in contrast to that observed for IC pigs fed 0 or 30% DDGS, or 30% DDGS diets with a 5 or 7 wk withdrawal before harvest, where overall ADG was not different among dietary feeding strategies (Asmus et al., 2014b). In the current study, the WD feeding strategy was not effective at improving overall ADG compared with pigs fed NCon. However, pigs fed SD tended ($P < 0.10$) to have greater overall ADG compared

with pigs fed NCon (0.94 vs. 0.91 ± 0.03 kg, respectively). In fact, overall ADG of pigs fed SD was similar to pigs fed PCon (0.94 vs. 0.96 ± 0.03 kg, respectively). This also resulted in similar final BW between pigs fed SD and PCon (123.1 vs. 125.3 ± 0.09 kg, respectively; Table 2.9).

Each TD treatment had a unique ADG pattern over time ($P < 0.01$; Figure 2.4 and Appendix Table A.5). Average daily gain of TD9 pigs peaked during the 15 to 17 wk interval, plateaued ($P > 0.05$) from 17 to 21 WOA, and then decreased ($P < 0.05$) during the 21 to 24 wk interval. This response is in contrast to that of TD7 pigs, where ADG peaked during the 19 to 21 wk interval, and then declined ($P < 0.05$). For TD5 pigs, ADG peaked at the 19 to 21 wk interval and remained constant ($P > 0.05$) to the end of the trial. These time-course changes resulted in reduced ($P < 0.05$) ADG in TD7 pigs compared with TD9 pigs during the 15 to 17 wk interval (0.93 vs. 1.06 ± 0.03 kg, respectively). During the 21 to 24 wk interval, TD5 pigs had greater ($P < 0.05$) ADG than TD9 pigs (1.07 vs. 0.93 ± 0.03 kg, respectively). However, these changes in ADG among TD treatments did not result in changes in BW over time (Table 2.10 and Appendix Table A.5). The changes observed in ADG, and lack of change in BW among TD treatments, may be explained by changes in body composition. This is important to consider given the advantage of greater lean gain of pigs prior to the second Improvest® (Squires, 2011) and the inherent increase in backfat deposition after the second Improvest® dose (Lealiifano et al., 2011). Changes in body composition of IC pigs fed different DDGS feeding strategies will be discussed in Chapter 4.

E. Gain efficiency

Gain efficiency of pigs fed PCon was improved ($P < 0.05$) compared with pigs fed SD and WD, and tended ($P < 0.10$) to be improved compared with pigs fed NCon during phase 1 (Figure 2.5 and Appendix Table A.3). This contributed to an overall improvement ($P < 0.05$) in gain efficiency of pigs fed PCon and SD compared with pigs fed WD and NCon (0.427 and 0.424 vs. 0.414 and 0.413 ± 0.005, respectively; Table 2.8). Bergstrom et al. (2009) reported that pigs fed 60% DDGS diets had poorer feed efficiency compared with pigs fed 20% DDGS diets. In contrast, when feeding diets containing up to 45% DDGS, gain efficiency was similar across DDGS diet inclusion levels (Cromwell et al., 2011). However, gain efficiency varied among collaborating geographical locations (Cromwell et al., 2011). Each location supplied their own corn and soybean meal but used the same source of DDGS. This suggests that factors other than DDGS nutrient compositional differences may contribute to the variable growth performance responses summarized by Stein and Shurson (2009).

Compared with PC, IC pigs have improved overall gain efficiency (Asmus et al., 2014b, Puls et al., 2014), and feed efficiency (Pauly et al., 2009, Batorek et al., 2012). In the current study, gain efficiency of TD9 pigs was poorer ($P < 0.05$) than TD7 pigs during the 17 to 19 wk interval, and gain efficiency of TD9 and TD7 pigs was poorer ($P < 0.05$) during the 19 to 21 wk interval (Figure 2.5 and Appendix Table A.2). It is well established that gain efficiency is reduced with advancing age of pigs. Interestingly, the 2 wk time intervals before and after the second Improvest® dose were not different for each TD treatment, which prolonged improved gain efficiency. Prolonging gain efficiency improvements among TD over time resulted in an overall improvement ($P <$

0.05) of gain efficiency in TD5 pigs compared with TD9 pigs, and a tendency for improved ($P < 0.05$) gain efficiency compared with TD7 pigs (Table 2.8). While TD9 pigs had poorer gain efficiency compared with TD5 pigs in this study, it is unknown how these responses may compare with those of PC because PC were not evaluated in this study. However, other researchers have reported that relative to other studies, TD9 pigs had improved overall gain efficiency compared with PC pigs (Puls et al., 2014; Asmus et al., 2014b). Therefore, harvesting pigs 9 wk after the second Improvest® dose may still capture improvements in gain efficiency relative to PC pigs. In fact, Lealiifano et al. (2011) observed similar gain efficiency when increasing the interval between the second Improvest® dose and harvest from 2 to 6 wk.

In summary, there were no 3-way interactions among FS, TD, and wk for any measure of growth performance. Feed intake of IC pigs fed 40% DDGS diets may have been limited due to the high fiber content and the age at when the second Improvest® dose was administered. The withdrawal of DDGS from the diet 5 wk before harvest resulted in a rapid increase of ADFI. These changes in ADFI likely resulted in changes in energy and nutrient intake. Overall, the SD feeding strategy was slightly more effective than the WD feeding strategy in maintaining ADFI, ADG, and gain efficiency similar to pigs fed PCon. Overall gain efficiency was improved in TD5 pigs compared with TD7 and TD9 pigs, but the gain efficiency observed for TD7 and TD9 was improved compared with results from what other studies for PC pigs.

Figure 2.1. Schematic of experimental design, sequence of dietary changes, and timing of Improvest® administration

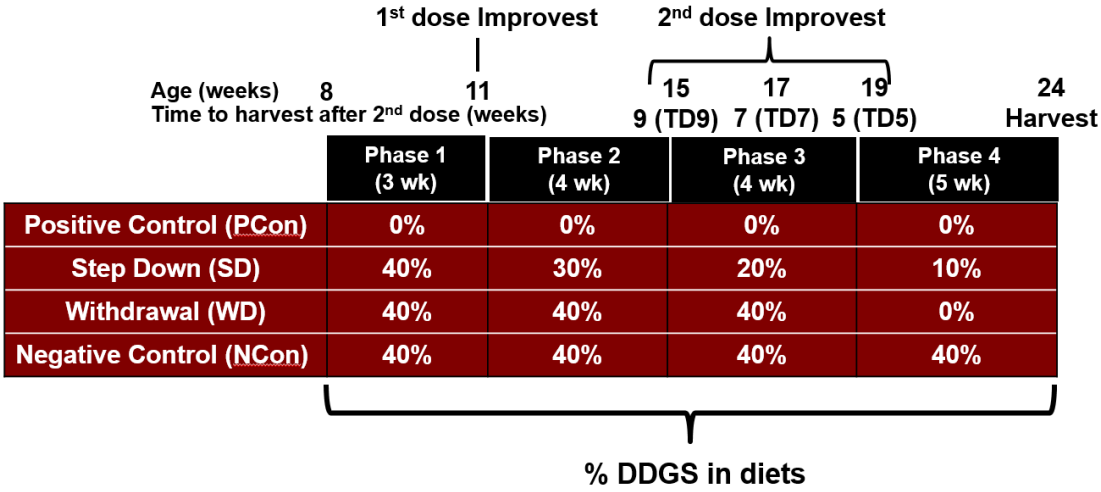


Table 2.1. Diet composition and calculated energy and nutrient composition of phase 1 and 2 diets

	Phase 1		Phase 2		
Ingredient, %	0% DDGS	40% DDGS	0% DDGS	30% DDGS	40% DDGS
Corn	61.23	36.23	69.03	49.22	41.06
Soybean meal (47.5% CP)	35.94	20.70	28.15	17.80	16.10
Corn DDGS[†]	0.00	40.00	0.00	30.00	40.00
0% DDGS basemix[1]	2.73	0.75	2.65	1.20	0.50
40% DDGS basemix[2]	0.00	1.95	0.00	1.44	2.00
L-lysine HCl	0.00	0.27	0.07	0.24	0.24
Lincomycin[3]	0.10	0.10	0.10	0.10	0.10
Calculated composition[4]					
ME, kcal/kg	3,322	3,219	3,329	3,250	3,228
NE, kcal/kg	2,202	2,209	2,235	2,235	2,231
CP, %	22.2	23.6	19.2	20.6	21.8
Ether extract, %	3.5	6.0	3.6	5.4	6.0
Total Lys, %	1.24	1.26	1.08	1.10	1.11
SID Lys, %	1.10	1.07	0.96	0.94	0.93
SID Met, %	0.31	0.35	0.28	0.31	0.33
SID Thr, %	0.72	0.72	0.62	0.63	0.66
SID Trp, %	0.24	0.20	0.20	0.17	0.18
Ca, %	0.69	0.75	0.65	0.70	0.70
Available P, %	0.32	0.37	0.30	0.35	0.35
SID Lys: ME, g/Mcal	3.31	3.32	2.88	2.89	2.88
IVP[5]	37.1	65.8	40.0	61.1	67.6

[†] DDGS = dried distillers grains with solubles.

[1]0% DDGS Basemix = 41.35% monocalcium phosphate, 34.40% limestone, 10.95% salt, 8.75% vitamin trace mineral premix, 3.55% corn; 1.0% soybean oil.

[2]40% DDGS Basemix = 62.25% monocalcium phosphate, 11.10% limestone, 10.70% vitamin trace mineral premix, 10.65% salt, 4.30% corn; 1.0% soybean oil.

[3]Linco20 (Lincomycin Hydrochloride; Zoetis, Inc. Florham Park, N.J., 07932).

[4] (NSNG, 2010)

[5] Calculated using values obtained from analyzed lipid content and fatty acid composition; Iodine value product (IVP) = (iodine value of lipid) × (ether extract, %) × 0.10 (Madsen et al., 1992), where iodine value of lipid = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723 (AOCS, 1998).

Table 2.2. Diet composition and calculated energy and nutrient composition of phase 3 and phase 4 diets

	Phase 3			Phase 4		
Ingredient, %	0% DDGS	20% DDGS	40% DDGS	0% DDGS	10% DDGS	40% DDGS
Corn	74.88	60.70	43.87	77.03	70.84	45.37
Soybean meal (47.5% CP)	22.20	16.50	13.50	20.00	16.20	12.05
Corn DDGS[†]	0.00	20.00	40.00	0.00	10.00	40.00
0% DDGS basemix[1]	2.70	1.50	0.28	2.75	2.10	0.25
40% DDGS basemix[2]	0.00	1.00	2.06	0.00	0.57	2.05
L-lysine HCl	0.12	0.20	0.19	0.12	0.19	0.18
Lincomycin[3]	0.10	0.10	0.10	0.10	0.10	0.10
Calculated composition[4]						
ME, kcal/kg	3,329	3,283	3,232	3,333	3,309	3,239
NE, kcal/kg	2,258	2,258	2,244	2,266	2,269	2,251
CP, %	16.9	18.3	20.7	16.0	16.4	20.2
Ether extract, %	3.6	4.8	6.0	3.6	4.3	6.0
Total Lys, %	0.96	0.98	1.00	0.89	0.91	0.95
SID Lys, %	0.85	0.84	0.83	0.79	0.79	0.78
SID Met, %	0.25	0.28	0.31	0.24	0.25	0.31
SID Thr, %	0.54	0.56	0.62	0.51	0.50	0.60
SID Trp, %	0.17	0.16	0.16	0.16	0.15	0.15
Ca, %	0.64	0.64	0.67	0.64	0.65	0.65
Available P, %	0.30	0.31	0.33	0.30	0.30	0.32
SID Lys: ME, g/Mcal	2.55	2.56	2.55	2.38	2.38	2.41
IVP[5]	42.0	55.8	68.6	42.8	50.0	69.1

[†] DDGS = dried distillers grains with solubles.

[1]0% DDGS Basemix = 41.35% monocalcium phosphate, 34.40% limestone, 10.95% salt, 8.75% vitamin trace mineral premix, 3.55% corn; 1.0% soybean oil.

[2]40% DDGS Basemix = 62.25% monocalcium phosphate, 11.10% limestone, 10.70% vitamin trace mineral premix, 10.65% salt, 4.30% corn; 1.0% soybean oil.

[3]Linco20 (Lincomycin Hydrochloride; Zoetis, Inc.Florham Park, N.J., 07932).

[4] (NSNG, 2010)

[5] Calculated using values obtained from analyzed lipid content and fatty acid composition; Iodine value product (IVP) = (iodine value of lipid) × (ether extract, %) × 0.10 (Madsen et al., 1992), where iodine value of lipid = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723 (AOCS, 1998).

Table 2.3. Energy and nutrient values of corn, soybean meal, and dried distillers grains with solubles (DDGS) used for diet formulation

Item	Corn[1]	Soybean Meal[1]	DDGS[2]
DM, %	88.3	90.0	88.6
ME, kcal/kg	3,395	3,294	3,144
CP, %	8.2	47.7	26.2
Crude fat, %	3.5	1.5	9.7
ADF, %	2.9	5.3	12.1
NDF, %	9.1	8.2	29.0
SID Lysine, %	0.19	2.63	0.55
SID Threonine, %	0.22	1.58	0.76
SID Methionine, %	0.15	0.59	0.41
SID Tryptophan, %	0.05	0.60	0.15
P_{avail}, %	0.05	0.15	0.54

[1] (NRC, 2012).
[2] Illuminate® (Value Added Science and Technology, Mason City IA).

Table 2.4. Analyzed fatty acid content of corn dried distillers grains with solubles (DDGS)

Fatty acid[1]	% of ether extract
C14:0 Myristic acid	0.06
C16:0 Palmitic acid	15.07
C17:0 Margaric acid	0.08
C18:0 Stearic acid	2.03
C20:0 Arachidic acid	0.40
C22:0 Behenoic acid	0.24
C24:0 Lignoceric acid	0.28
C16:1-n9 Palmitoleic acid	0.14
C17:1-n10 Heptadecenoic acid	0.04
C18:1-n9c Oleic acid	24.96
C18:1-n9t Elaidic acid	0.07
C20:1-n9 Gonodic acid	0.45
C18:2-n6 Linoleic acid	53.90
C18:3-n3 Linolenic acid	1.71
C22:6-n3 Docosahexaenoic acid	0.21
Total SFA[2]	18.16
Total MUFA[3]	25.21
Total PUFA[4]	55.82

Table 2.5. Analyzed nutrient composition of corn dried distillers grains with solubles (DDGS), as-fed basis

Item	Analyzed composition
DM, %	86.7
CP, %	28.6
Ether extract, %	10.4
Ash, %	3.9
ADF, %	12.3
NDF, %	34.1
Ca, %	0.04
P, %	0.79
IVP[1]	124.5
Indispensable AA, %	
Arg	1.31
His	0.78
Ile	1.06
Leu	3.45
Lys	0.99
Met	0.57
Phe	1.42
Thr	1.13
Trp	0.21
Val	1.45
Dispensable AA, %	
Ala	1.96
Asp	1.78
Cys	0.51
Glu	3.53
Gly	1.16
Pro	2.35
Ser	1.31
Tyr	1.16

[1] Calculated using values obtained from analyzed lipid content and fatty acid composition; Iodine value product (IVP) = (Iodine value of fat) × (ether extract, %) × 0.10 (Madsen et al., 1992), where Iodine value of fat = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723 (AOCS, 1998).

Table 2.6. Analyzed composition of phase 1 and phase 2 diets, as-fed basis[1]

	Phase 1		Phase 2		
Nutrient composition	0% DDGS	40% DDGS	0% DDGS	30% DDGS	40% DDGS
CP, %	20.92	22.84	17.56	20.10	19.77
Ether extract, %	1.97	5.04	2.04	4.06	5.06
ADF, %	3.57	6.67	3.55	4.88	5.09
NDF, %	9.22	16.77	9.50	14.24	15.57
Lys, %	1.21	1.21	1.00	1.08	1.13
Met, %	0.30	0.37	0.27	0.34	0.37
Thr, %	0.77	0.87	0.64	0.73	0.79
Trp, %	0.25	0.24	0.19	0.19	0.22
Ca, %	0.91	0.89	0.76	0.80	0.77
P, %	0.69	0.69	0.60	0.63	0.62
IVP[2]	23.9	60.5	25.6	49.5	61.2
Fatty acids[3,4]					
C16:0 Palmitic acid	14.94	15.18	14.20	14.63	14.95
C18:0 Stearic acid	2.41	2.09	2.24	2.11	2.17
Other SFA[5]	0.69	0.59	0.56	0.64	0.76
C16:1n9 Palmitoleic acid	0.11	0.15	0.14	0.14	0.18
C18:1n9 Oleic acid	21.71	23.38	23.42	24.51	24.48
C18:2n6 Linoleic acid	55.15	53.92	55.22	54.53	53.84
C18:3n3 Linolenic acid	4.31	2.35	3.62	2.31	2.36
C20:1n9 Gonodic acid	0.00	0.36	0.00	0.44	0.33
C20:5n3 Eicosapentaenoic acid	0.00	0.00	0.00	0.00	0.03
C22:6 n3 Docosahexaenoic acid	0.00	0.07	0.00	0.09	0.11
Total SFA[6]	18.51	18.20	17.32	17.80	18.02
Total MUFA[7]	21.81	23.54	23.56	24.64	24.72
Total PUFA[8]	59.46	56.37	58.83	56.93	56.34

[1]Each value represents the mean of 4 diet mixing lots.

[2] Calculated using values obtained from analyzed lipid content and fatty acid composition; Iodine value product (IVP) = (Iodine value of fat) × (crude fat, %) × 0.10, where Iodine value of fat = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723.
[3]Expressed as a percentage of total ether extract.
[4]Diets contained no detectable levels of the following fatty acids: myristoleic acid (C14:1n9), pentadecanoic acid (C15:0), C17:1n10, elaidic acid (C18:1n9t), vaccenic acid (C18:1n11), stearidonic acid (C18:4n3), homo-α-linolenic acid (C20:3n3), arachidonic acid (C20:4n6), 3n-arachidonic acid (C20:4n3), erucic acid (C22:1n9), clupanodonic acid (C22:5n3), nervonic acid (C24:1n9).
[5]Other SFA = myristic acid (C14:0), margaric acid (C17:0), behenoic acid (C22:0), lignoceric acid (C24:0).
[6] Total SFA = myristic acid (C14:0), palmitic acid (C16:0), margaric acid (C17:0) stearic acid (C18:0), arachidic acid (C20:0), behenoic acid (C22:0), and lignoceric acid (C24:0).
[7] Total MUFA = palmitoleic acid (C16:1n-9), oleic acid (C18:1n-9c), and gonodic acid (C20:1n-9).
[8] Total PUFA = linoleic aicd (C18:2n-6) and linolenic acid (C18:3n-3).

Table 2.7. Analyzed nutrient composition of phase 3 and phase 4 diets[1]

	Phase 3			Phase 4		
Nutrient composition, %	0% DDGS	20% DDGS	40% DDGS	0% DDGS	10% DDGS	40% DDGS
CP	16.19	17.56	20.29	15.18	15.39	19.58
Ether extract	2.10	3.45	4.89	2.23	3.06	5.46
ADF	3.32	4.16	5.81	3.26	3.98	5.92
NDF	9.67	13.68	17.99	10.01	11.22	16.73
Lys	0.94	0.95	1.03	0.84	0.91	0.96
Met	0.26	0.29	0.38	0.24	0.27	0.35
Thr	0.57	0.64	0.75	0.54	0.57	0.72
Trp	0.19	0.21	0.20	0.20	0.18	0.19
Ca	0.63	0.77	0.76	0.64	0.71	0.74
P	0.58	0.57	0.63	0.50	0.55	0.61
IVP[2]	26.0	42.1	59.1	27.4	36.7	65.8
Fatty acids[3,4], %						
C16:0 Palmitic acid	13.88	14.71	15.00	13.90	15.00	14.96
C18:0 Stearic acid	2.30	2.20	2.10	2.27	2.20	2.05
Other SFA[5]	0.63	0.62	0.71	0.50	0.53	0.60
C16:1n9 Palmitoleic acid	0.10	0.14	0.14	0.17	0.23	0.14
C18:1n9 Oleic acid	24.47	24.37	24.60	24.77	23.79	24.75
C18:2n6 Linoleic acid	54.77	54.49	54.22	53.95	53.44	54.03
C18:3n3 Linolenic acid	3.03	2.43	2.06	2.92	2.47	1.99
C20:1n9 Gonodic acid	0.22	0.43	0.45	0.29	0.32	0.42
C20:5n3 Eicosapentaenoic acid	0.00	0.00	0.00	0.00	0.00	0.00
C22:6 n3 Docosahexaenoic acid	0.00	0.06	0.09	0.00	0.00	0.06
Total SFA[6]	17.16	17.91	18.20	16.90	17.68	18.00
Total MUFA[7]	24.57	24.50	24.78	24.93	24.02	24.93
Total PUFA[8]	57.79	56.97	56.37	56.87	55.91	56.11

[1]Each value represents the mean of 4 diet mixing lots.

[2] Calculated using values obtained from analyzed lipid content and fatty acid composition; Iodine value product (IVP) = (Iodine value of fat) × (crude fat, %) x 0.10, where Iodine value of fat = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723.
[3]Expressed as a percentage of total ether extract.
[4]Diets contained no detectable concentrations of the following fatty acids: myristoleic acid (C14:1n9), pentadecanoic acid (C15:0), C17:1n10, elaidic acid (C18:1n9t), vaccenic acid (C18:1n11), stearidonic acid (C18:4n3), homo-α-linolenic acid (C20:3n3), arachidonic acid (C20:4n6), 3n-arachidonic acid (C20:4n3), erucic acid (C22:1n9), clupanodonic acid (C22:5n3), nervonic acid (C24:1n9).
[5]Other SFA = myristic acid (C14:0), margaric acid (C17:0), behenoic acid (C22:0), lignoceric acid (C24:0).
[6] Total SFA = myristic acid (C14:0), palmitic acid (C16:0), margaric acid (C17:0), stearic acid (C18:0), arachidic acid (C20:0), behenoic acid (C22:0), and lignoceric acid (C24:0).
[7] Total MUFA = palmitoleic acid (C16:1n-9), oleic acid (C18:1n-9c), and gonodic acid (C20:1n-9).
[8] Total PUFA = linoleic aicd (C18:2n-6) and linolenic acid (C18:3n-3).

Table 2.8. Overall (16 wk) growth performance of immunologically castrated pigs using different DDGS feeding strategies (FS) at 3 different time intervals between the second Improvest® dose and harvest.

	Feeding Strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD[3]
ADFI, kg/head/d	2.396^{x}	2.345^{xy}	2.392^{xy}	2.339^{y}	0.075	2.399	2.392	2.313	0.078	0.04	0.16	0.07
ADG, kg/head/d	0.96^{aw}	0.94^{aby}	0.93^{bcx}	0.91^{cz}	0.03	0.93	0.94	0.94	0.03	< 0.01	0.77	0.77
G:F	0.427^{a}	0.424^{a}	0.414^{b}	0.413^{b}	0.005	0.413^{a}	0.417^{ax}	0.428^{by}	0.005	< 0.01	0.03	0.37

[1]PCon = pigs fed corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = pigs fed diets containing 40, 30, 20, and 10% DDGS in the 4 dietary phases, respectively; WD = pigs fed diets containing 40% DDGS in phases 1to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wks of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[3] See Figure 2.2 and Appendix Table A.1 for interactive means.

[a,b,c] Within a row of main effects FS and TD, means without a common superscript differ ($P \leq 0.05$).

[w,x] Within a row of main effects FS and TD, means without a common superscript differ ($P \leq 0.10$).

[y,z] Within a row of main effects FS and TD, means without a common superscript differ ($P \leq 0.10$).

Table 2.9. Interactive BW least squares means at the beginning of each dietary phase, and final BW of immunologically castrated pigs when fed different dried distillers grains with solubles DDGS feeding strategies (FS) and harvested at 3 different time intervals after the second Improvest® dose (See also Appendix Table A.4)

Trait	Feeding strategies[1,2,3]			
Initial BW, kg	PCon	SD	WD	NCon
Phase 1	21.5	21.5	21.5	21.5
Phase 2	36.8	35.6	35.4	35.6
Phase 3	62.2[a]	60.1[ab]	59.0[b]	59.3[b]
Phase 4	91.7[a]	88.6[b]	86.8[b]	86.4[b]
Final BW, kg	125.3[a]	123.1[ab]	122.3[bc]	120.0[c]

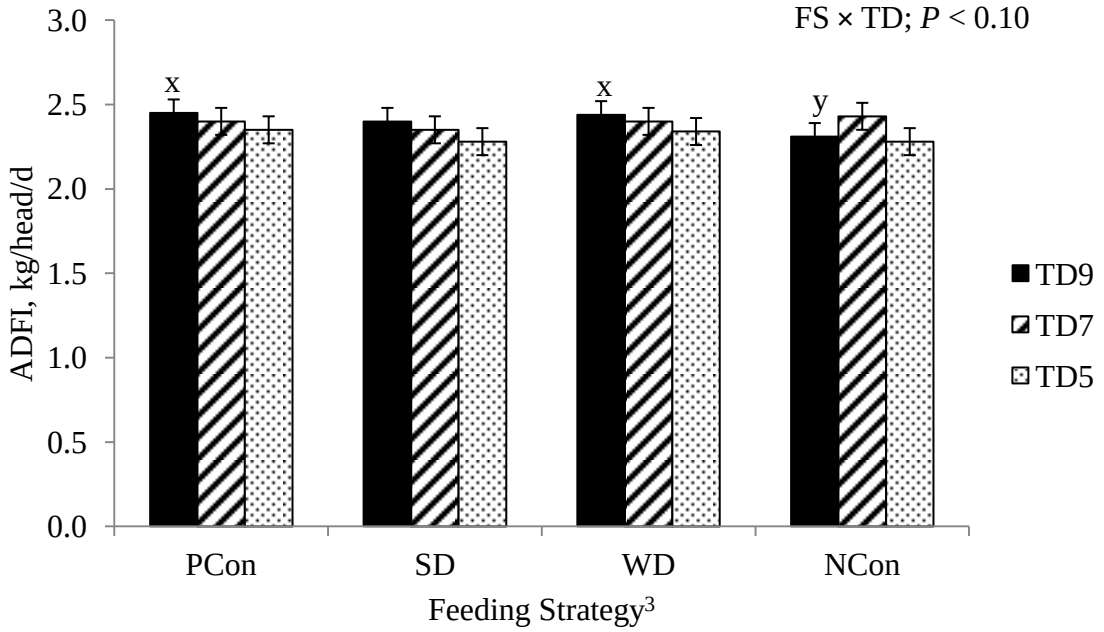
[1] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout the growing-finishing period.
[2] FS × TD × Wk; and FS × TD ($P \geq 0.21$).
[3] FS × Wk ($P < 0.001$); SEM 0.9 kg.
[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).

Table 2.10. Interactive BW least squares means at the beginning of each dietary phase, and final BW of immunologically castrated pigs when fed different dried distillers grains with solubles feeding strategies (FS) and harvested at 3 different time intervals after the second Improvest® dose (see also Appendix Table A.5)

Trait	Interval between second Improvest® dose and harvest (TD)[1,2,3]		
Initial BW, kg	TD9	TD7	TD5
Phase 1	21.5	21.4	21.5
Phase 2	35.8	35.8	35.9
Phase 3	59.7	60.2	60.5
Phase 4	89.2	88.0	87.9
Final BW, kg	121.9	122.7	123.4

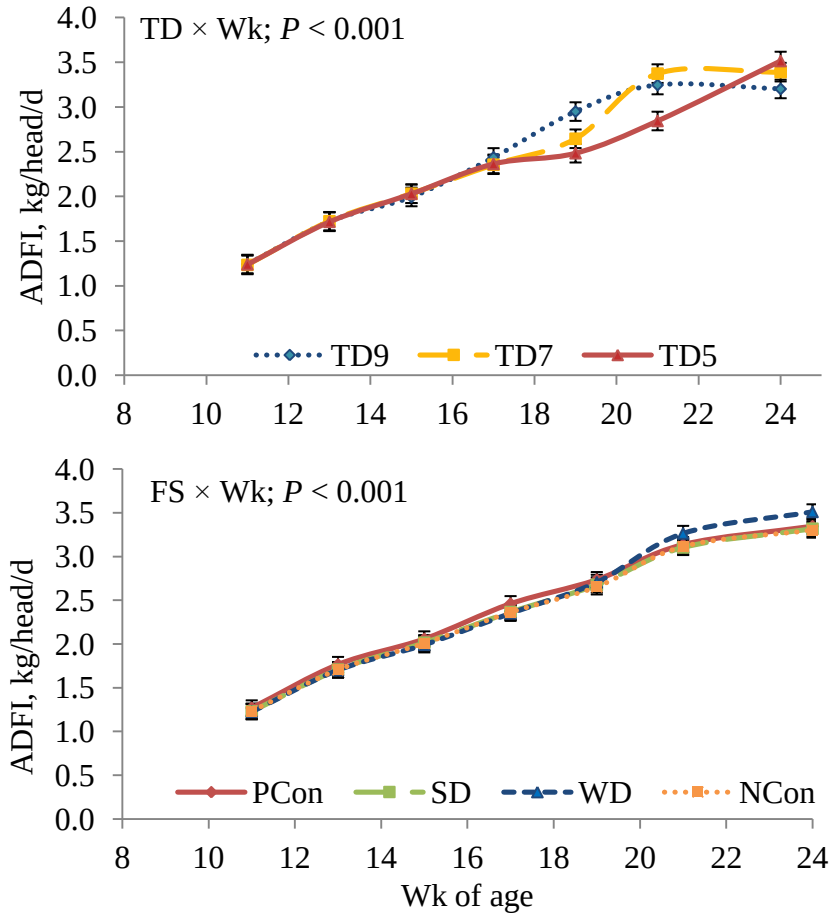
[1] All pigs received the first dose of Improvest® at 11 wks of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] FS × TD × wk; and FS × TD ($P \geq 0.21$).
[3] TD × Wk ($P = 0.99$); SEM 1.2 kg.

Figure 2.2. Overall ADFI of immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 dried distillers grains with solubles (DDGS) feeding strategies (FS)[1,2]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] See Appendix Table A.1 for least squares means.
[3] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout the growing-finishing period.
[x,y] Means without a common superscript differ ($P \leq 0.10$).

Figure 2.3. Average daily feed intake of immunologically castrated pigs with increasing time intervals between the second Improvest® dose and harvest and fed differing dried distillers grains with solubles (DDGS) feed strategies (FS) from 8 to 24 weeks of age (See also Appendix Tables A.2 and A.3)



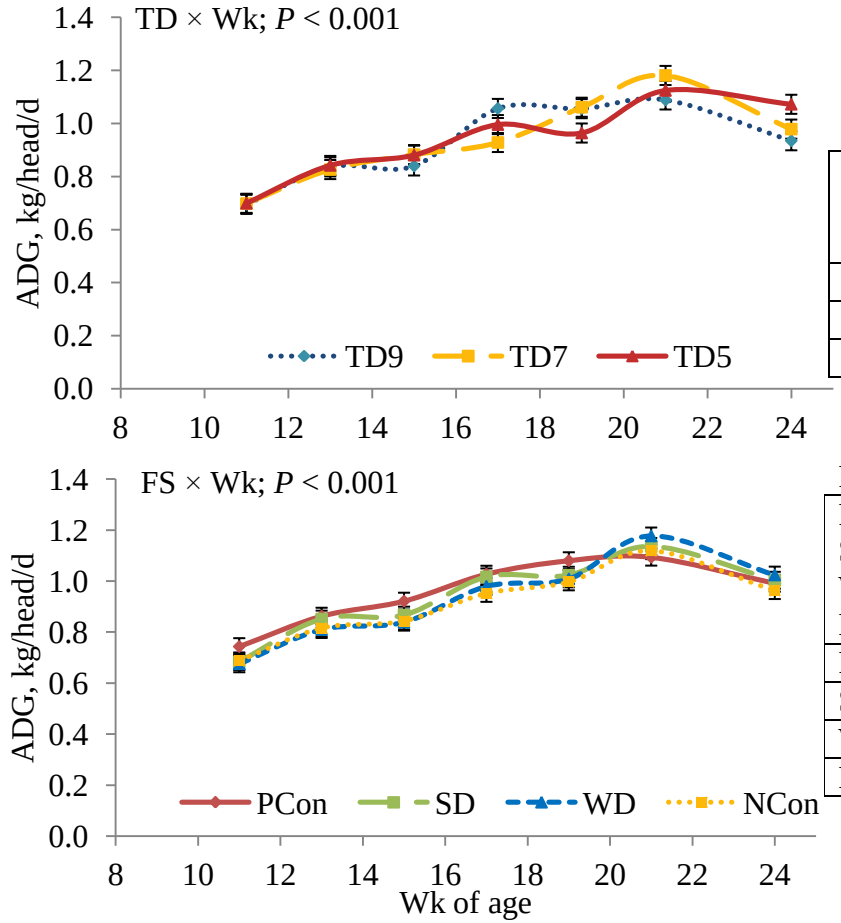
Time interval between 2nd Improvest® dose and harvest[1]	Wk of age						
	11	13	15	17	19	21	24
TD9					a	a	
TD7					ab	a	
TD5					b	b	
TD9	A	B	B	C	D	D	D
TD7	A	B	BC	CD	D	E	E
TD5	A	B	BC	CD	DE	E	F

FS[2]	Wk of age						
	11	13	15	17	19	21	24
PCon						ab	b
SD						b	b
WD						a	a
NCon						b	b
PCon	A	B	C	D	E	F	F
SD	A	B	C	D	E	F	F
WD	A	B	C	D	E	EX	Y
NCon	A	B	C	D	E	F	F

[1] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout the growing-finishing period.
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[A,B,C,D,E,F] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 2.4. Average daily gain of immunologically castrated pigs with increasing time intervals between the second Improvest® dose and harvest and fed dried distillers grains with solubles (DDGS) feed strategies (FS) from 8 to 24 wk of age (See also Appendix Tables A.4 and A.5)



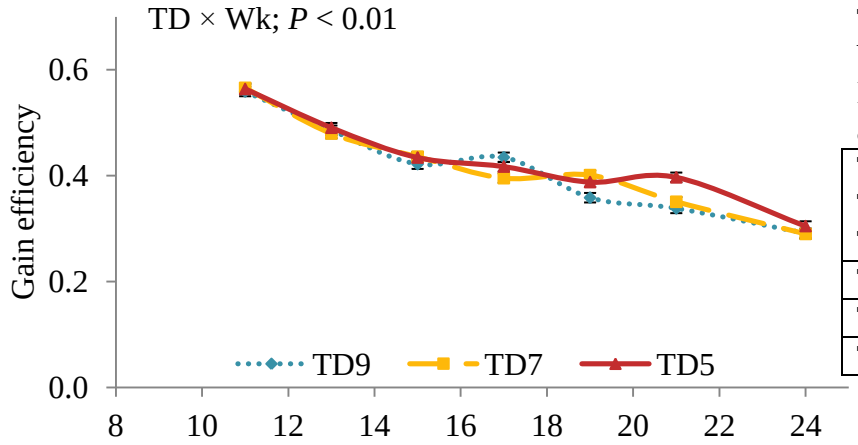
Time interval between 2nd Improvest® dose and harvest[1]	Wk of age						
	11	13	15	17	19	21	24
TD9				a			a
TD7				b			ab
TD5				ab			b
TD9	A	B	B	CY	CY	C	BX
TD7	A	B	B	D	C	E	CD
TD5	A	BX	BC	CD	CDY	E	E

FS[2]	Wk of age						
	11	13	15	17	19	21	24
PCon			ax	x	a	a	
SD			ab		ab	ab	
WD			b		ab	b	
NCon			y	y	b	ab	
PCon	A	B	BC	DE	DE	EY	CDX
SD	A	B	B	C	C	D	C
WD	A	B	B	C	C	D	C
NCon	A	B	B	C	C	D	C

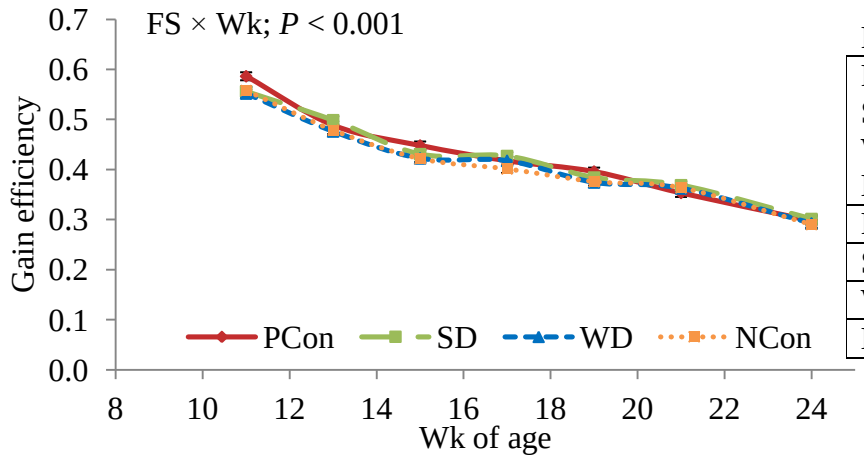
[1] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout the growing-finishing period.
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[A,B,C,D,E,F] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 2.5. Gain efficiency of immunologically castrated pigs with increasing time intervals between the second Improvest® dose and harvest and fed dried distillers grains with solubles (DDGS) feed strategies (FS) from 8 to 24 weeks of age (See also Appendix Tables A.2 and A.3)



Time interval between 2nd Improvest® dose and harvest[1]	Wk of age						
	11	13	15	17	19	21	24
TD9					b	b	
TD7					a	b	
TD5					ab	a	
TD9	A	B	C	C	D	D	E
TD7	A	B	CX	CY	C	D	E
TD5	A	B	C	CD	D	CD	E



FS[2]	Wk of age						
	11	13	15	17	19	21	24
PCon	ax						
SD	b						
WD	b						
NCon	y						
PCon	A	B	C	CD	D	E	F
SD	A	B	C	C	D	D	E
WD	A	B	C	C	D	D	E
NCon	A	B	C	CD	DE	E	F

[1]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout the growing-finishing period.
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

CHAPTER 3: Energy and nutrient intake and dietary cost of lean gain of immunologically castrated pigs harvested at 5, 7, or 9 weeks after the second Improvest® dose and under varying feeding strategies using corn dried distillers with solubles

I. Summary

The objective of this study was to determine the effects of using different dried distillers grains with solubles (**DDGS**) feeding strategies (**FS**) and increasing the time interval between the second Improvest® (*gonadotropin releasing factor (GnRF) analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest (**TD**) of IC pigs on energy and nutrient intake and dietary cost of lean gain. Intact male (**IM**) pigs (n = 863) were assigned randomly in a 4 × 3 factorial arrangement of treatments at 8 wk of age (**WOA**). Pigs were fed 1 of 4 DDGS FS in a 4-phase feeding program consisting of 3, 4, 4, and 5 wk for phases 1 to 4, respectively. Feeding strategies were: 1) corn-soybean meal (**CS**) control diet (**PCon**), 2) CS + 40% DDGS (**NCon**), 3) CS + 40, 30, 20, and 10% DDGS for phases 1 to 4, respectively (**SD**) or 4) CS + 40% DDGS in phase 1 to 3 and CS in phase 4 (**WD**). Pigs received the second Improvest® dose at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest. Differences in ME and nutrient intake among FS were due to differences in feed intake. Overall ME intake was reduced ($P < 0.05$) in pigs fed NCon compared with pigs fed PCon and WD (7.56 vs. 7.98 and 7.82 ± 0.25 Mcal/d, respectively), which was primarily due to lower ($P < 0.05$) overall ME intake of TD9 pigs fed NCon compared with TD9 pigs fed PCon and WD (7.48 vs. 8.15 and 7.97 ± 0.27 Mcal/d, respectively). Due to the higher CP content (28.6%) relative to lysine content in DDGS, pigs fed NCon had greater ($P < 0.05$) standardized ileal digestible (SID) CP

intake compared with all other feeding strategies, especially for TD5 and TD7 pigs fed NCon compared with TD5 and TD7 pigs fed PCon and SD. Similar SID Lys intake was observed among FS for TD5 and TD7 pigs, but TD9 pigs fed NCon had lower ($P < 0.05$) SID Lys intake compared with TD9 pigs fed PCon, and tended ($P < 0.10$) to have lower SID Lys intake compared with pigs fed WD (19.89 vs. 21.44 and 21.01 ± 0.66 g/d, respectively). Feeding NCon resulted in poorer ($P < 0.05$) lean gain ME efficiency (kg carcass lean/Mcal intake) compared with feeding SD (0.0401 vs. 0.0428 ± 0.0010 kg/Mcal, respectively). Caloric efficiency of TD5 pigs was improved ($P < 0.05$), and TD5 pigs had less ($P < 0.05$) backfat compared with TD9 and TD7 pigs. While lean gain/d was similar among IC pigs, TD5 pigs had improved ($P < 0.05$) lean gain ME efficiency compared with TD9 and TD7 pigs (0.0427 vs. 0.0409 and 0.0408 ± 0.0010 kg/Mcal, respectively). Compared with the PCon feeding strategy, using the SD feeding strategy was more effective at reducing the dietary cost of lean gain than using the WD feeding strategy.

KEYWORDS: dried distillers grains with solubles, energy, feeding strategy, immunological castration, lean gain efficiency, pigs

II. Introduction

Feed cost represents the largest proportion (54 to 78%) of total cost of producing pork (Schulz, 2014). Use of less expensive feed ingredients such as dried distillers grains with solubles (**DDGS**; Woyengo et al., 2014), and partitioning nutrients towards lean tissue accretion and away from fat accretion (van Milgen and Noblet, 2003) can reduce the cost of pork production. Immunological castration (**IC**) captures the inherent advantage of greater lean growth of intact male (**IM**) pigs compared with physical

castrates (**PC**), while minimizing the unpalatable off-odors of boar taint in meat products from IM pigs (Dunshea et al., 2001). However, increasing the interval between the second Improvest® (*gonadotropin releasing factor (**GnRF**) analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest results in progressively greater ADFI (Elsbernd et al., 2014).

Corn DDGS has become a common ingredient fed to pigs, but diets containing 30 to 45% DDGS can reduce feed intake, carcass dressing percentage, and carcass fat firmness in PC and gilts (Xu et al., 2010b; Cromwell et al., 2011; Graham et al., 2014). These negative effects can be overcome by using feeding strategies (**FS**) such as withdrawing (**WD**) DDGS from the diet (Gaines et al., 2007; Xu et al., 2010a) or gradually decreasing (**SD**) the DDGS inclusion throughout the growing-finishing period. Corn DDGS is a good source of energy, and digestible AA and P, but has a relatively high fiber content and poor protein quality due to its relatively low Lys relative to CP content (Stein and Shurson, 2009). The energetic efficiency of CP and fiber is poorer than fat and starch, and can limit the energy available for lean and adipose tissue deposition (Patience, 2012). Therefore, understanding the changes in energy and nutrient intake resulting from changes in feed intake patterns of IC pigs is important for determining optimal DDGS feeding strategies to optimize lean growth rate and efficiency, and reduce the dietary cost of lean gain.

III. Materials and methods

A. Animals and treatments

A detailed description of the experimental procedures used in this study is described in Chapter 2. Briefly, 4 groups of IM pigs (n = 863; initial BW = 21.5 ± 0.8 kg;

V100 Landrace females x V40 Large White boars; Genetiporc, Alexandria, MN) were assigned randomly to one of 12 treatments in a 4 × 3 factorial arrangement with 4 feeding strategies and 3 Improvest® treatments.

Each of the 4 feeding strategies utilized 4 phases where phases 1 to 4 were fed for 3, 4, 4, and 5 wk, respectively. Feeding strategy treatments included: a positive control (**PCon**) strategy where pigs were fed a corn-soybean meal based diet containing no DDGS throughout the growing-finishing period; a DDGS step down (**SD**) strategy where pigs were fed 40, 30, 20, and 10% DDGS in the 4 dietary phases, respectively; a DDGS withdrawal (**WD**) strategy where pigs were fed 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; and a negative control (**NCon**) strategy where pigs were fed 40% DDGS diets throughout the entire growing-finishing period (Chapter 2, Figure 2.1 and Tables 2.1 and 2.2). All pigs had ad libitum access to feed and water.

The first dose of Improvest® was administered to all pigs at 11 wk of age (**WOA**). The second dose of Improvest® was administered at 15, 17, or 19 WOA, which corresponded with 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest, respectively. Two wk after the second Improvest® dose, a quality assurance assessment was conducting by individually observing pigs for aggressive boar-like behavior, testes size, and color of the scrotum (FSIS, 2013). Pigs not meeting the quality assurance specifications (n = 38) received a third Improvest® dose and remained on the study.

B. Diet formulation and composition

A detailed description of diet composition and nutrient analysis is described in Chapter 2. All diets were formulated to meet or exceed energy and nutrient requirements for growing IM pigs (NRC, 2012). Dietary concentrations for standardized ileal

digestible (SID) Lys, as well as Ca, and P were increased by 5% as a safety margin to assure that these nutrients would not limit the growth of IM pigs. All diets were formulated to have similar SID Lys:ME and Ca:P within each phase (Table 2.1 and 2.2). Dried distillers grains with solubles was obtained from a single source and contained 10.4% ether extract (Table 2.4 and 2.5). Estimates of ME, SID AA, and available P content of this DDGS source were obtained from Illuminate® (Value Added Science and Technology, Mason City, IA) and used in diet formulation. Estimates of ME, SID AA, and available P content for corn and soybean meal were obtained from (NRC, 2012).

C. Energy and nutrient intake

Feed disappearance was determined at the beginning and end of each dietary phase, and 2 wk after the beginning of phases 2 to 4. The feed remaining at the end of each feeding period was subtracted from the total feed offered throughout each feeding period to determine feed disappearance. Energy and nutrient intake was calculated for each feeding period where ME intake = (total feed intake, kg × calculated Mcal of ME/kg of the diets)/d on feed; SID CP intake = (total feed intake × calculated SID CP % of the diets)/d on feed; SID Lys intake = (total feed intake × calculated SID Lys % of the diets)/d on feed, and ME efficiency = (total Mcal of ME intake/ total BW gain).

D. Ultrasound and carcass characteristics

Ultrasound (Aloka 500V SSD; Hitachi-Aloka Medical Ltd., Tokyo, Japan) evaluation for 10^{th} rib backfat (BF) and LM area was conducted on all pigs by a single trained and certified technician 4 d before harvest. At harvest, HCW for each pig was obtained and recorded. From these data, carcass dressing percentage was calculated by (HCW, kg/final BW, kg) × 100, and percentage fat-free lean was calculated according to

NPPC (2000) where percentage carcass fat-free lean = ((final carcass fat-free lean / [HCW, kg]) × 100). Final carcass fat-free lean was calculated by [2.620 + (0.401 × HCW, kg) – (3.358 × ultrasound 10^{th} rib backfat depth, cm) + (0.306 × ultrasound 10^{th} rib LM, cm^2) + (0.456 × sex of the pig; barrow=1, gilt=2)]. Lean gain per day was calculated as (final carcass fat-free lean – initial fat-free lean)/number of days on feed; where initial fat-free lean (NPPC, 2000) = [(0.922 × initial BW, kg) – 3.65]. Lean gain efficiency was calculated for each pen as (final carcass fat-free lean – initial fat-free lean)/total feed intake.

E. Statistical analysis

Energy and nutrient intake data were analyzed using PROC MIXED in SAS (Cary, NC) where pen was considered the experimental unit. The statistical model included feeding strategy, TD, and feeding strategy × TD as fixed effects, and group as a random effect. Week was used as a repeated measure. During lairage, 1 pig died and 15 carcasses required primal trimming. These pigs were removed from the dataset for analysis of carcass characteristic data. Hot carcass weight was used as a covariate ($P < 0.01$) for final BF and LM area ultrasound data analysis. The residuals were used to test the assumptions of normality using PROC Univariate. When the Kolmogorov-Smirnov test for normality was significant ($P < 0.05$), any datapoint with a residual > ± 4 was removed from the analysis. This included 3 data points for ME intake and 1 data point for final ultrasound BF and carcass dressing percentage. Least squares means were separated and adjusted using the Tukey option. Significance was determined when $P \leq 0.05$ and trends were identified when $P \geq 0.05$ and ≤ 0.10.

IV. Results and discussion

A. Metabolizable energy intake

Typically, voluntary feed intake of growing-finishing pigs, when feed is provided *ad libitum*, is dependent on dietary energy density so that energy intake is similar regardless of feed intake (Ellis and Augsperger, 2001). In this study, diets were not isocaloric. Therefore, ADFI should have been different among feeding strategies to achieve similar energy intake. However, the differences in ADFI for various DDGS FS (described in Chapter 2) did not result in pigs with similar ME intake (Figure 3.1 and Appendix Table A.6) as expected. Instead, pigs consuming a lower ME density diet also consumed less feed. Pigs fed NCon had lower ($P < 0.05$) overall ME intake than pigs fed PCon and WD (7.56 vs. 7.98 and 7.82 ± 0.25 Mcal/d, respectively; Table 3.1). In addition, pigs fed SD had lower ($P < 0.05$) ME intake than pigs fed PCon, but similar ME intake compared with pigs fed NCon (7.69 vs. 7.98 and 7.56 Mcal/d ± 0.25, respectively). This was primarily due to TD9 pigs fed PCon and WD having greater ($P < 0.05$) ME intake than TD9 pigs fed NCon (8.15 and 7.97 vs. 7.48 ± 0.27, respectively; Figure 3.1 and Appendix Table A.6). Moreover, TD5 pigs fed SD, and TD9 and TD5 pigs fed NCon had lower ($P < 0.05$) ME intake than TD9 pigs fed PCon (7.49, 7.48, and 7.37 vs. 8.15 ± 0.27 Mcal/d, respectively). Therefore, differences in ME intake are a result of the time of administering the second Improvest® dose before harvest and the feeding strategy used.

While there was no TS × FS × wk interaction for ME intake, the overall differences in ME intake can be attributed to TD × wk and FS × wk interactions during the growing-finishing period. Reduction of ME intake began during the 13 to 15 wk interval. Pigs fed PCon tended ($P < 0.10$) to have greater ME intake compared with pigs

fed WD (6.85 vs. 6.43 ± 0.29 Mcal/d, respectively; Figure 3.2 and Appendix Table A.7). This continued into phase 3 where during the 15 to 17 wk interval, pigs fed PCon had greater ($P < 0.05$) ME intake compared with pigs fed WD and NCon (8.19 vs. 7.60 and 7.63 ± 0.29 Mcal/d, respectively) and pigs fed PCon tended ($P < 0.10$) to have greater ME intake compared with pigs fed SD (7.76 ± 0.29 Mcal/d). It is possible that feed intake of pigs fed NCon and WD was limited by the greater fiber content of the diets containing 40% DDGS preventing pigs from achieving a similar ME intake. Others have reported decreased ADFI when feeding diets greater than 40% but have not calculated ME intake. Feeding high fiber diets can result in greater intestinal fill due to the greater bulk density and increased water retention of fiber (Kerr and Shurson, 2013). Abruptly reducing the DDGS inclusion from 40% to 0% would allow for greater potential to increase ME intake if the diet has lower fiber content. This physical effect of fiber may explain why ADFI and ME intake increased even though pigs fed WD were switched to a higher ME density diet (Tables 2.1 and 2.2), which should have resulted in reduced ADFI.

Immunological castrates did not increase ADFI to account for lower dietary ME density. This was observed by the lack of changes in ADFI before the 19 to 21 wk interval, and the varying ME intake beginning in the 13 to 15 wk interval. During the 17 to 19 wk and 19 to 21 wk intervals, TD5 pigs had lower ($P < 0.05$) ME intake than TD9 pigs (8.12 vs. 9.64 and 9.40 vs. 10.72 ± 0.34 Mcal/d, respectively; Figure 3.2 and Appendix Table A.8). After the TD5 pigs received the second Improvest® dose at 19 WOA, like ADFI, ME intake increased ($P < 0.05$) rapidly from the 19 to 21 wk compared with the 21 to 24 wk intervals (9.40 vs. 11.61 ± 0.34 Mcal/d). During this same time, ME intake of TD7 and TD9 pigs remained unchanged. There are many physiological factors

that regulate feed intake in addition to diet ME content (Black et al., 2009). In this case, the suppression of steroid hormones that occurs shortly after the second dose of Improvest® is administered (Claus et al., 2007) appears to have a greater influence on regulating feed intake than the energy density of the diet. In addition, greater consumption of a higher energy density diet along with the suppression of anabolic hormones would suggest that a greater proportion of ME intake would be partitioned toward adipose tissue accretion and away from lean tissue accretion resulting in reduced lean gain efficiency after the second Improvest® dose. As expected, TD7 pigs had improved ($P < 0.05$) ME efficiency compared with TD9 pigs during the 17 to 19 wk interval (8.19 vs. 9.16 ± 0.34, respectively) since TD9 pigs received the second Improvest® dose at 15 WOA, which was 2 wk earlier than TD7 pigs. During the 19 to 21 wk interval, TD5 pigs had improved ($P < 0.05$) ME efficiency compared with TD9 and TD7 pigs (8.36 vs. 9.80 and 9.46, respectively), since TD9 and TD7 pigs received the second Improvest® dose 4 and 2 wk prior to the 19 to 21 wk interval. These results indicate that immunological castration changes the ME intake and ME efficiency pattern of pigs after the second Improvest® dose. Thus, it would be appropriate in practice to provide pigs with a diet that reflects the androgenic-induced shift in ME intake.

Based on the hormonal suppression that occurred during the 10 d period after the second Improvest® dose, and greater feed intake of IC pigs compared with PC within 14 d after the second Improvest® dose (Claus et al., 2007), there may be some residual lean growth advantages realized by delaying dietary changes until 7 to 14 d after the second Improvest dose. This concept is supported by the recommendation of Dunshea et al. (2013) that IC pigs should be fed a PC diet beginning 1 wk after the second Improvest®

dose. These researchers also suggested that the SID Lys requirement of IC pigs at 4 to 5 wk after the second Improvest® dose should be 6% below the SID Lys requirement of gilts (Dunshea et al., 2013). However, given the continual increase in ADFI for at least 4 (Elsbernd et al., 2014) or 6 wk (Lealiifano et al., 2011) after the second Improvest® dose, use of a customized diet for a second IC dietary phase may be beneficial for optimizing dietary energy and AA utilization. Since ME intake of pigs in this study did not follow the assumption that pigs would voluntarily adjust ADFI and achieve similar ME intake, this can result in lower intake of other nutrients such as AA.

B. Nutrient intake

In this study, the DDGS source contained 28.6% CP. While DDGS contains a relatively high concentration of CP, it is a poor quality protein source because of its relatively low Lys content. In swine diets, Lys is the first limiting AA and the dietary adequacy of all other AA are in proportion to Lys. Amino acids fed in excess of these proportions are not utilized for protein accretion but are deaminated for use as energy sources, which is an energetically expensive process and increases N excretion (Le Bellego et al., 2001).

Standardized ileal digestibility (**SID**) of AA is the proportion of AA of greatest interest because SID corrects for the AA that will be not be digested regardless of dietary ingredients (basal endogenous AA losses; Stein et al., 2007) and reflects the AA that the pig can use for protein deposition. Thus, the resulting diet SID values reflect digestibility differences among different feed ingredients that comprise the diet (Stein et al., 2007). There were no FS × TD × wk interactions for SID CP or SID Lys intake. The time interval between the second Improvest® dose and harvest did not alter SID CP or SID

Lys intake beyond the changes observed for ADFI until the 21 to 24 wk interval, when intake of SID CP (Figure 3.4 and Appendix Table A.10) and SID Lys (Figure 3.6 and Appendix Table A.10) were greater ($P < 0.05$) in TD5 pigs compared with TD9. Overall, SID Lys intake was reduced ($P < 0.05$) in pigs fed NCon compared with pigs fed PCon (20.14 vs. 21.05 ± 0.63, respectively; Table 3.1), which was due to TD9 pigs fed NCon having reduced SID Lys intake compared with TD9 pigs fed PCon, and a trend ($P < 0.10$) for TD9 pigs fed NCon to have lower SID Lys intake compared with TD9 pigs fed WD (19.89 vs. 21.44 and 21.01 ± 0.66 g/d, respectively; Figure 3.5 and Appendix Table A.6). All other TD treatments had similar SID Lys intake among FS. Standardized ileal digestible Lys intake was similar among FS over time until the final 3 wk period (Figure 3.6 and Appendix Table A.9). The increase of ADFI during the final 3 wk period of pigs fed WD also led to an increase in SID Lys intake, where in the final 3 wk, pigs fed WD had greater ($P < 0.05$) SID Lys intake compared with pigs fed NCon (27.75 vs. 25.97 ± 0.69 g/d, respectively). Intake of SID Lys in pigs fed WD was similar to pigs fed PCon and SD during the 21 to 24 wk interval (27.75 vs. 26.45 and 26.48 ± 0.69 g/d, respectively).

The high CP intake can be attributed to the dietary feed ingredients, feeding strategies, and the formulation of diets to meet requirements for IM pigs in this study. Overall, SID CP intake was much greater ($P < 0.05$) in pigs fed NCon and WD compared with pigs fed PCon and SD (388.6 and 374.3 vs. 357.1 and 353.7 ± 0.63 g/d, respectively; Table 3.1). This increase was due to the increased SID CP intake of pigs fed NCon and WD compared with pigs fed PCon and SD beginning in the 15 to 17 wk (385.4 and 387.3 vs. 353.6 and 352.4 ± 12.4 g/d, respectively) and 17 to 19 wk (443.3 and 494.6 vs. 393.1

and 398.8 ± 12.4 g/d, respectively) intervals (Figure 3.4 and Appendix Table A.9). Furthermore, the high CP content of NCon and WD diets containing 40% DDGS caused CP intake to be greater compared with other diets and feeding strategies. Balancing the diets on a NE basis would have accounted for the energetic expense of metabolizing CP because 1.86 kJ/g of diet goes toward heat production (Noblet and van Milgen, 2013). As a result, CP on an ME basis has an energy value of 103% of starch, but on an NE basis is only 71% the energy value of starch (Noblet and van Milgen, 2013). Thus, the efficiency of energy utilization from CP is poor (Noblet and van Milgen, 2013). The addition of crystalline Lys would have improved the overall protein quality of the diets and allowed more of the excess amino acids present to be utilized for protein accretion. Even with an improvement of protein quality, adequate energy intake must be provided for optimal protein accretion (van Milgen and Noblet, 2003). Energy intake may have been a key limiting factor resulting from the high fiber content of DDGS and the ADFI limitation due to physical gut capacity. Energy intake could have been corrected with the addition of another source of supplemental fat, but this would have confounded the additional study objectives of determining pork fat quality and feeding strategies to overcome the effects of feeding diets high in unsaturated fatty acid content (e.g. DDGS) that have been shown consistently to reduce pork fat quality. Energy supplementation must coincide with an improvement in protein quality to improve lean accretion, otherwise excess energy is deposited as adipose tissue.

As previously discussed, feeding high CP diets decreases the energy utilization efficiency on an ME basis. In the present study, all diets, including diets fed after the second dose of Improvest®, were formulated for IM pigs using a 5% safety margin for

SID Lys to assure that IM pigs were allowed to achieve their maximum lean growth potential. In addition, the Lys and CP were fed in excess of the estimated requirements during the time when feed intake increased due to IC, regardless of feeding strategy, resulting in greater CP consumption. Therefore, CP was fed in excess for longer periods of time as the time interval between second Improvest® and harvest increased. This may have resulted in a plateau in feed intake. Feeding of 13, 16, 19, 22, and 25% CP diets have been shown to linearly decrease feed intake, and tended to linearly decrease ADG (Chen et al., 1999). Phase 4 diets fed in the current study contained 15.18, 15.39, and 19.58% CP in 0, 10, 40% DDGS diets, respectively. While overall ADFI decreased linearly with increasing CP, due to excess nitrogen metabolism, a linear increase in liver and kidney weights has been observed (Chen et al., 1999). Both are major sites of AA degradation and nitrogen excretion. The increase in body weight and metabolic function increases the energy demand for maintenance of larger organs, resulting in greater heat production and less energy available for the pig to use for growth (Chen et al., 1999). Not only is excess protein not utilized for growth, but it increases nitrogen excretion and concentration of nitrogen in manure. This becomes an environmental concern because application rates of swine manure to cropland are limited by its nitrogen content. Pork production systems have overcome this challenge by formulating diets based on SID AA and improving the overall protein quality of diets by adding supplemental crystalline AA. The feeding of high levels of DDGS to IC pigs with increasing ADFI after the second Improvest® dose requires better understanding of the feed intake dynamics and nutrient requirements of IC pigs after the second Improvest® dose.

C. Hot carcass weight

As a result of reduced overall ADG, HCW of pigs fed NCon was less ($P < 0.05$) compared with pigs fed PCon (HCW: 85.3 vs. 90.6 ± 1.5 kg, respectively; Table 3.2). This is consistent with results from other studies where diets containing up to 40% (Graham et al., 2014), 45% (Cromwell et al., 2011), or 60% DDGS (Bergstrom et al., 2009a; Leick et al., 2010; Hardman, 2014) were fed. Hot carcass wt of pigs using the SD and WD feeding strategies resulted in a tendency ($P < 0.10$) for reduced HCW compared with pigs fed PCon (88.3 and 87.8 vs. 90.6 ± 1.5 kg, respectively).

All other studies that have evaluated carcass characteristics of IC pigs with increasing time intervals between the second Improvest® dose and harvest have increased the time interval by increasing the age of the pigs (Asmus et al., 2014b; Boler et al., 2014; Tavárez et al., 2014b). Boler et al. (2014) and Asmus et al. (2014) observed the increase in HCW with increasing age was similar in PC and IC pigs when harvested at 4 or 6 wk (Boler et al., 2012) and 5 or 7 wk after the second Improvest® dose (Asmus et al., 2014b). However, Tavárez et al. (2014) demonstrated that HCW of IC pigs was greater than PC pigs at 6 wk after the second Improvest® dose, and was not different between IC and PC pigs at 8 wk after the second Improvest® dose. In the present study, all pigs were harvested at 24 WOA, and increasing the interval between the second Improvest® dose and harvest from 5 to 9 wk did not affect final BW or HCW (Table 3.2).

D. Dressing percentage

Feeding diets containing DDGS has been reported to reduce dressing percentage when using dietary inclusion rates up to 30% (Whitney et al., 2006; Xu et al., 2010b;

Asmus et al., 2014b), 40% (Graham et al., 2014) or 60% (Leick et al., 2010; Hardman, 2014). Use of a WD feeding strategy to recover the loss in dressing percentage has been effective for improving the dressing percentage when DDGS was withdrawn 30 d before harvest (Hill et al., 2008) or 21 or 42 d before harvest (Gaines et al., 2007) in PC and gilts. In the present study, IC pigs fed NCon had lower ($P < 0.05$) dressing percentage compared with pigs fed PCon. The expected losses of dressing percentage may have been due to the fiber-associated increase in visceral mass that occurs when feeding high fiber diets (Kerr and Shurson, 2013), such as DDGS, compared with feeding corn-soybean meal diets. In this study, the DDGS source contained 34.1% NDF and 12.3% ADF. The complete diets with 0% DDGS and diets with 40% DDGS contained 9.22% and 16.77% NDF, respectively in phase 1, 9.50% vs. 15.57% NDF, respectively in phase 2, 9.67% vs. 17.99% NDF, respectively in phase 3, and 10.01% vs. 16.73% NDF, respectively in phase 4. Other researchers have reported recently that decreasing the feeding duration of high fiber diets (19% NDF from DDGS and wheat middlings) linearly improved carcass dressing percentage over time (Asmus et al., 2014a). In the present study, the SD and WD strategies were effective in recovering the loss in dressing percentage resulting in pigs fed the PCon, SD, and WD strategies, resulting in similar dressing percentages (72.3, 71.8, and 72.0 ± 0.3%, respectively; Table 3.2).

Other researchers have consistently reported reduced dressing percentage of IC pigs compared with PC (Pauly et al., 2009; Gispert et al., 2010; Boler et al., 2012; Asmus et al., 2014b; Boler et al., 2014), but similar dressing percentage compared with IM pigs (Pauly et al., 2009; Boler et al., 2014). In addition to the more developed reproductive tract of IC pigs, the empty small intestine, stomach, blood (Boler et al., 2014), and liver

(Pauly et al., 2011; Boler et al., 2014) wt made up a greater proportion of the BW in IC than PC pigs. The increased visceral mass can be attributed to greater androgen receptors on visceral organs which are responsive to circulating testosterone leading to greater protein synthesis and weight gain (Wade and Gray, 1979) of IM and IC pigs before the second Improvest® dose. Therefore, it was not surprising that in the present study, delaying the second dose of Improvest® reduced ($P < 0.05$) carcass dressing percentage in TD5 pigs compared with TD7 pigs, and tended ($P < 0.10$) to reduce dressing percentage in TD9 pigs (71.4 vs. 72.0 and 72.0 ± 0.03%, respectively). Boler et al. (2012) did not observe this difference between pigs receiving the second Improvest® dose at 4 or 6 wk before harvest, but the pigs receiving the second Improvest® does at 6 wk were harvested 2 wk later than pigs receiving the second dose at 4 wk before harvest.

E. Ultrasound backfat and LM area

The proportion of lean and adipose accretion is the result of genetic potential for lean gain and nutrient intake to meet the lean gain potential (van Milgen and Noblet, 2003). Individually, feeding DDGS diets to pigs and use of immunological castration have resulted in variable changes in carcass composition. Most body composition evaluations of pigs fed DDGS diets or for IC pigs have occurred in the carcass (discussed in Chapter 5), and few studies have evaluated body composition using live animal ultrasound. Hardman (2014) reported a linear decrease in live animal ultrasound of LM depth, but observed no change in BF thickness when feeding diets containing up to 60% DDGS. In the present study, DDGS feeding strategy did not affect LM area. However TD5 pigs had reduced ($P < 0.05$) BF thickness compared with TD7 pigs (Table 3.2) due to TD7 pigs fed PCon and NCon having greater ($P < 0.05$) BF thickness than TD5 pigs

fed PCon and NCon feeding strategies (Figure 3.8). Other researchers have observed greater LM area in IC pigs compared with PC pigs, especially at 8 wk after the second Improvest® dose. Compared with IC pigs, PC pigs have greater BF thickness, and BF thickness increased with greater time intervals between the second Improvest® dose and harvest (Tavárez et al., 2014b).

F. Lean gain efficiency and dietary cost of lean gain

Although overall ME intake (Table 3.1) and final BW (Table 3.2) were not different among TD treatments, overall ME efficiency (Table 3.1) was improved ($P < 0.05$) in TD5 pigs compared with TD7 and TD9 pigs (7.92 vs. 8.20 and 8.31 ± 0.10, respectively). Moreover, final BW, HCW, LM area, and lean gain/d were not different among TD treatments, but since TD5 pigs consumed less feed and ME than TD7 and TD9 pigs, lean gain efficiency (0.141 vs. 0.134 and 0.135 ± 0.003 kg/kg, respectively) and lean gain ME efficiency of TD5 pigs was improved ($P < 0.05$) compared with TD7 and TD9 pigs.

Final BW was reduced due to reduced ($P < 0.05$) lean gain of pigs fed NCon compared with all other feeding strategies. The consumption of SID Lys was in excess of the NRC (2012) requirements for IM pigs and likely was not a limiting factor for lean gain. Therefore, it is likely that energy intake was a limiting factor in lean gain. When considering lean gain per Mcal of ME, pigs fed NCon also had poorer ($P < 0.05$) lean gain efficiency compared with pigs fed PCon and SD (0.0401 vs. 0.0420 and 0.0428 ± 0.0010 kg/Mcal, respectively). In addition, lean gain ME efficiency was similar among pigs fed PCon, SD, and WD, but pigs fed WD had poorer ($P < 0.05$) lean gain ME efficiency than pigs fed SD (0.0408 vs. 0.0428 ± 0.0010 kg/Mcal, respectively). Since

body wt gain ME efficiency was not different among FS, but lean gain efficiency was different, and there were no appreciable differences in BF or LM area, it is plausible that energy was partitioned toward other energy consuming processes such as toward meeting the greater maintenance energy needs of the larger visceral mass. As previously described, excess CP intake is energetically costly due to increased cost of urea production and increased organ mass, resulting in greater maintenance energy requirements. Consequently, this energy is diverted away from energy for lean or adipose tissue growth. Larger organs also contribute consistently to reduced dressing percentage in IC pigs compared with PC. Increasing the time interval between second Improvest® dose and harvest has not resulted in improving carcass dressing percentage at least up to 6 weeks (Lealiifano et al., 2011; Boler et al., 2012). In the current study, pigs receiving the second dose of Improvest® 5 weeks before harvest had the most appropriately matched diets because they were IM pigs for the longest period of time. This resulted in TD5 pigs having greater lean gain ME efficiency compared with TD7 and TD9 pigs.

Depending on the price relationship between DDGS, corn, and soybean meal, including DDGS in swine diets can reduce dietary cost (Woyengo et al., 2014). However, the decision to add DDGS to the diet must be considered in relation to lean gain/d and lean gain efficiency per unit of energy intake. The cost per kg of diet for each dietary phase of NCon was lower than the other FS (Table 3.3), it appears that pigs did not have sufficient energy intake to maintain similar lean gain/d as pigs fed the other FS (Table 3.2). Pigs fed the WD strategy consumed the same diets as pigs fed NCon until phase 4, at which time the removal of the DDGS from the diet resulted in a rapid increase in feed (and ME) intake of a more expensive diet (Table 3.3). This occurred at a time when lean

gain efficiency would inherently be lowest and therefore, add to the dietary cost of lean gain. The improvement in lean gain of pigs fed the SD and WD feeding strategies resulted in lower dietary cost of production compared with pigs fed NCon (Figure 3.9). In addition, pigs fed SD were able to maintain lean gain efficiency similar to pigs fed PCon, and pigs fed SD had improved lean gain efficiency compared with pigs fed the WD strategy. Moreover, pigs fed WD had a rapid increase in feed intake once the DDGS was removed during phase 4, resulting in pigs consuming more of a more expensive diet during phase 4 which increased the dietary cost of lean gain compared with feeding the SD strategy. Since the SD strategy diets were less expensive than PCon, and lean gain/d and lean gain efficiency were similar to pigs fed PCon, the dietary cost of lean gain was substantially reduced by using the SD feeding strategy. As the diet comprises the greatest production expense of pork production, the SD feeding strategy has the greatest economic benefit for reducing the cost of lean gain for IC pigs.

Great emphasis is placed on management strategies to reduce the cost of lean gain in pork production systems because of consumer preference for lean meat, packer carcass pricing incentives, and reduced dietary cost of depositing lean versus fat. Energy intake of IC pigs fed 40% DDGS may have been limited due to the high fiber content and the age at when the second Improvest® dose was administered in TD9 pigs. To overcome this, energy density would need to be increased using a supplemental fat source as well as using greater amounts of digestible AA to ensure that additional energy is partitioned toward lean deposition, and reduce the amount of excess CP in the diets. Dietary adjustments need to be made 10 d after the second Improvest® dose based on delayed suppression of anabolic hormones and the progressive ADFI increase. These dietary

adjustments could accommodate typical marketing strategies in the U.S., where removing the heaviest pigs from pens in 2 or 3 sequential marketings would inherently change the interval between second Improvest® dose and harvest. Additional dietary phase changes may be necessary to account for increased feed intake and nutrient intake beyond the second Improvest® dose to match the changing nutrient requirements of pigs. Since lean gain and lean gain efficiency were similar between pigs fed PCon and SD, the lower dietary cost of the SD strategy reduced the dietary cost of lean gain relative to pigs fed PCon. The SD strategy was more effective than the WD strategy in reducing the dietary cost of lean gain.

Table 3.1. Overall (16 wk) energy and nutrient intake of immunologically castrated pigs using different DDGS feeding strategies at 3 different time intervals between the second Improvest® dose and harvest(See also Figure 3.1 and Appendix Table A.6).

	Feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD
ME intake[3], Mcal/head/d	7.98[a]	7.69[bc]	7.82[ab]	7.56[c]	0.25	7.86	7.84	7.58	0.26	< 0.01	0.16	0.07
ME efficiency[4]	8.17	8.04	8.20	8.16	0.09	8.31[a]	8.20[a]	7.92[b]	0.10	0.14	0.02	0.26
SID CP intake[5], g/head/d	357.1[a]	353.7[a]	374.3[b]	388.6[c]	11.4	373.0[a]	371.8[ax]	360.5[by]	11.3	< 0.01	0.03	0.04
SID Lys intake[6], g/head/d	21.05[a]	20.41[b]	20.62[ab]	20.14[b]	0.63	20.82[a]	20.41[ax]	20.14[by]	0.63	< 0.01	0.03	0.07

[1]PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[3] ME intake calculated as (total feed intake × calculated dietary ME density)/ (pigs per pen × d on feed).

[4] ME efficiency calculated as total dietary ME intake/ total BW gain.

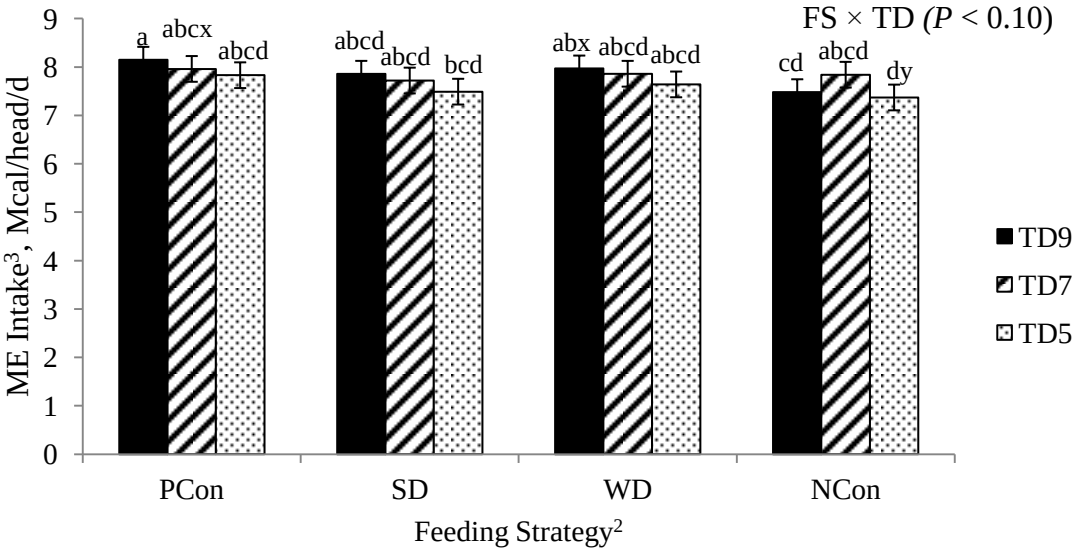
[5] SID CP intake calculated as (total feed intake × calculated dietary SID CP content)/(pigs per pen × d on feed).

[6] SID Lys intake calculated as (total feed intake × calculated dietary SID Lys content)/ (pigs per pen × d on feed).

[a,b,c] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.10$).

Figure 3.1. Overall ME intake of immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 dried distillers grains with solubles (DDGS) feeding strategies (FS; see also Appendix Table A.6)[1]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

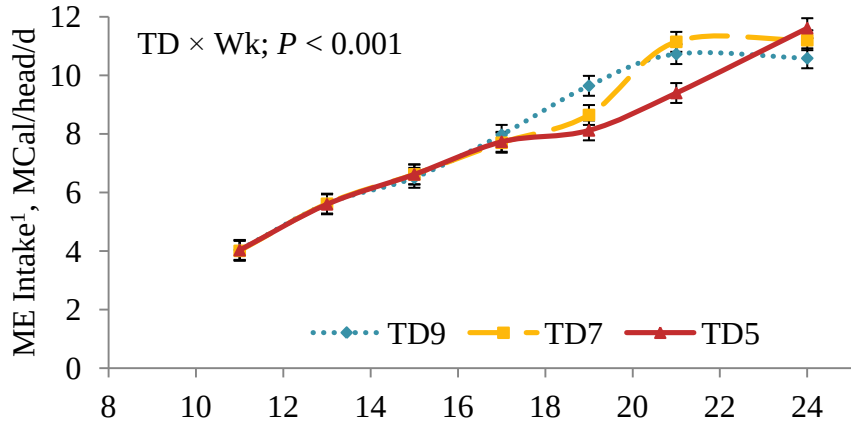
[2] PCon = pigs fed corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = pigs fed diets containing 40, 30, 20, and 10% DDGS in the 4 dietary phases, respectively; WD = pigs fed diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.

[3] For each weighed period, ME intake calculated as (total feed intake × calculated dietary ME density)/(pigs per pen × d on feed).

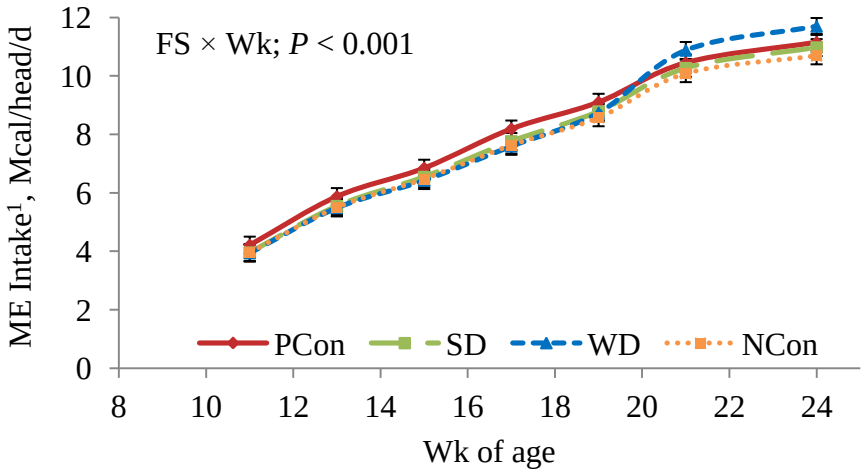
[a,b,c,d] Means without a common superscript differ ($P \leq 0.05$).

[x,y] Means without a common superscript differ ($P \leq 0.10$).

Figure 3.2. Metabolizable energy intake of immunologically castrated pigs fed dried distillers grains with solubles feeding strategies (FS) and harvested with increasing time intervals between the second Improvest® dose and harvest (See also Appendix Tables A.7 and A.8)



Time interval between 2nd Improvest® dose and harvest[2]	Wk of age						
	11	13	15	17	19	21	24
TD9					a	a	
TD7					ab	a	
TD5					b	b	
TD9	A	B	B	C	D	D	D
TD7	A	B	BC	CD	D	E	E
TD5	A	B	BC	CD	DX	EY	F

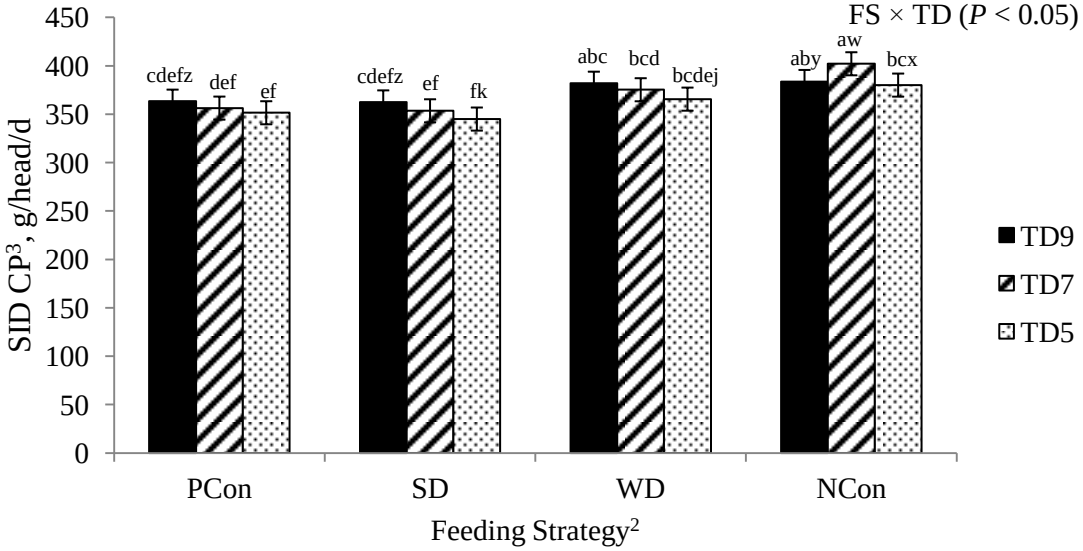


FS[3]	Wk of age						
	11	13	15	17	19	21	24
PCon			x	ax	a	bx	b
SD				by	ab	b	bc
WD			y	b	ab	ay	a
NCon				b	b	b	c
PCon	A	B	C	D	E	F	F
SD	A	B	C	D	E	F	F
WD	A	B	C	D	E	FX	Y
NCon	A	B	C	D	E	F	F

[1] ME intake calculated as (total feed intake × calculated dietary ME density)/(pigs per pen × d on feed).

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.
[3] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[a,b,c] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a column, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 3.3. Overall standardized ileal digestible (SID) CP intake of immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 dried distillers grains with solubles (DDGS) feeding strategies (FS; see also Appendix Table A.6)[1]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.

[3] For each weighed period, SID CP intake calculated as (total feed intake × calculated SID CP)/(pigs per pen × d on feed).

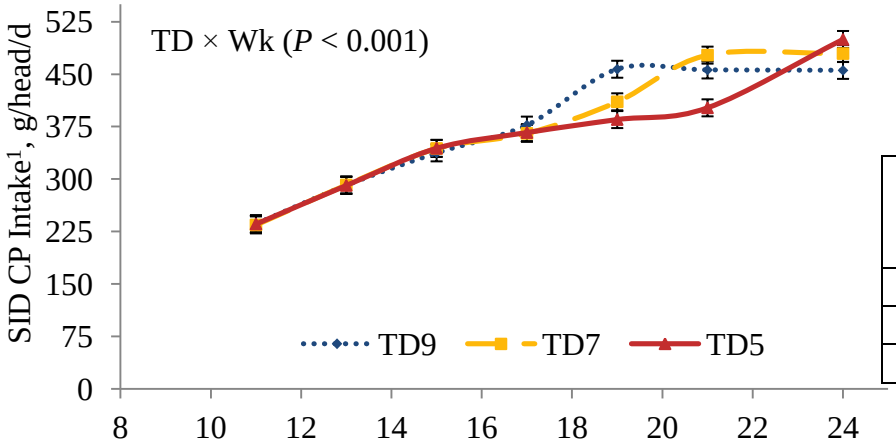
[a,b,c,d,e,f] Means without a common superscript differ ($P \leq 0.05$).

[w,x] Means without a common superscript differ ($P \leq 0.10$).

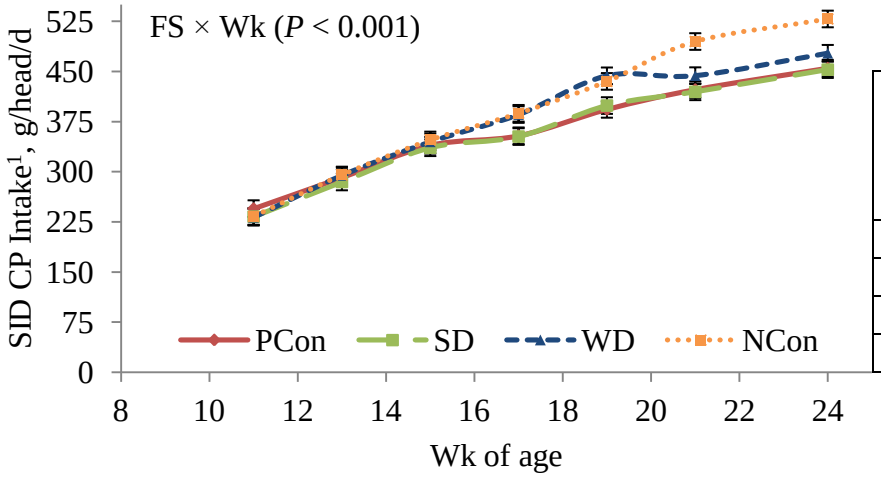
[y,z] Means without a common superscript differ ($P \leq 0.10$).

[j,k] Means without a common superscript differ ($P \leq 0.10$).

Figure 3.4. Standardized ileal digestible (SID) CP intake of immunologically castrated pigs fed dried distillers grains with solubles feeding strategies (FS) and harvested with increasing time intervals between the second Improvest® dose and harvest (See also Appendix Tables A.9 and A.10)



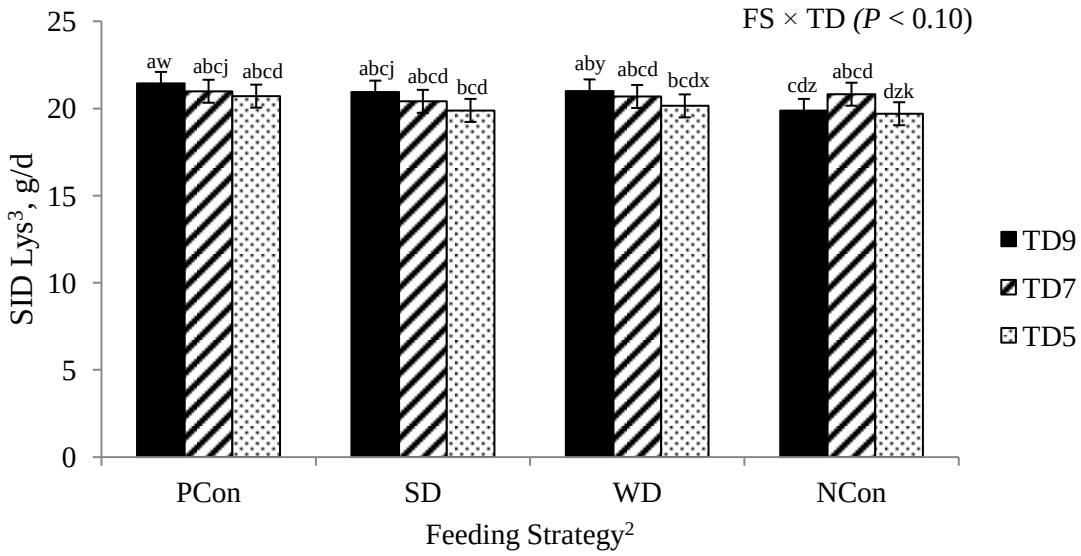
Time interval between 2nd Improvest® dose and harvest[2]	Wk of age						
	11	13	15	17	19	21	24
TD9					a	a	a
TD7					bx	a	ab
TD5					by	b	b
TD9	A	B	D	C	E	E	E
TD7	A	B	C	C	D	E	E
TD5	A	B	C	DX	D	EY	F



FS[3]	Wk of age						
	11	13	15	17	19	21	24
PCon				a	a	a	a
SD				a	a	a	a
WD				b	b	a	a
NCon				b	b	b	b
PCon	A	B	C	C	D	E	F
SD	A	B	C	C	D	D	E
WD	A	B	C	D	E	E	F
NCon	A	B	C	D	E	F	G

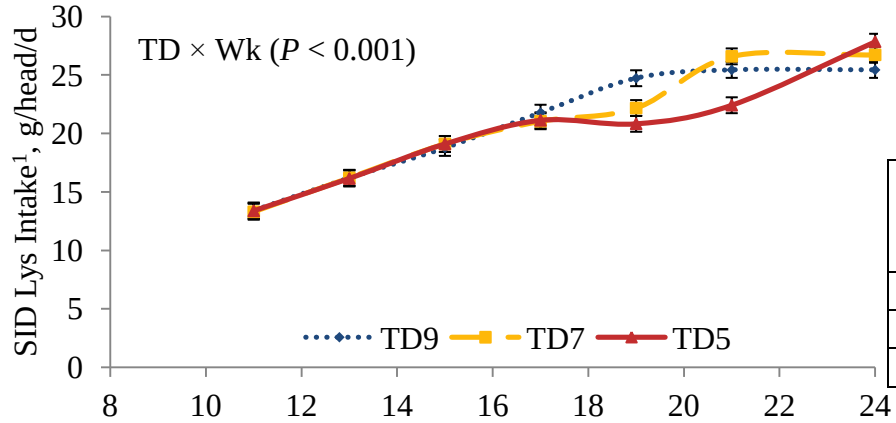
[1] For each feeding period, SID CP intake calculated as (total feed intake × calculated dietary SID CP content)/ (pigs per pen × d on feed).
[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.
[3] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a column, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F,G] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 3.5. Overall standardized ileal digestible (SID) Lys intake of immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 dried distillers grains with solubles (DDGS) feeding strategies (FS; see also Appendix Table A.6)[1]

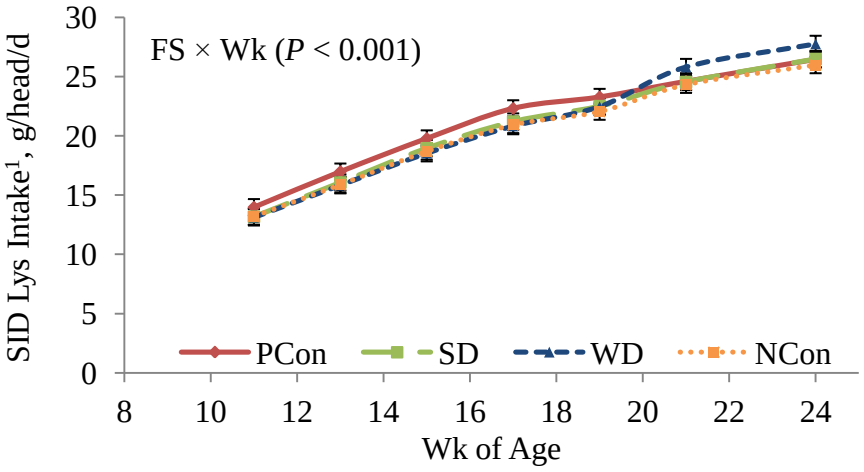


[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2]PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[3] For each weighed period, SID Lys intake calculated as (total feed intake × calculated SID Lys)/(pigs per pen × d on feed).
[a,b,c,d] Means without a common superscript differ ($P \leq 0.05$).
[w,x] Means without a common superscript differ ($P \leq 0.10$).
[y,z] Means without a common superscript differ ($P \leq 0.10$).
[j,k] Means without a common superscript differ ($P \leq 0.10$).

Figure 3.6. Standardized ileal digestible (SID) Lys intake of immunologically castrated pigs fed dried distillers grains with solubles feeding strategies (FS) and harvested with increasing time intervals between the second Improvest® dose and harvest (see also Appendix Table A.9 and A.10)



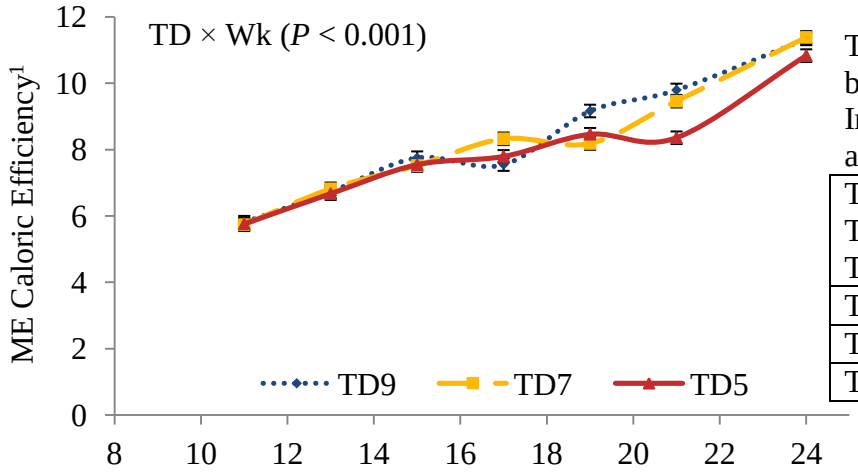
Time interval between 2nd Improvest® dose and harvest[2]	Wk of age 11	13	15	17	19	21	24
TD9					a	a	a
TD7					bx	a	ab
TD5					by	b	b
TD9	A	B	C	D	E	E	E
TD7	A	B	C	D	D	E	E
TD5	A	B	C	DX	D	EY	F



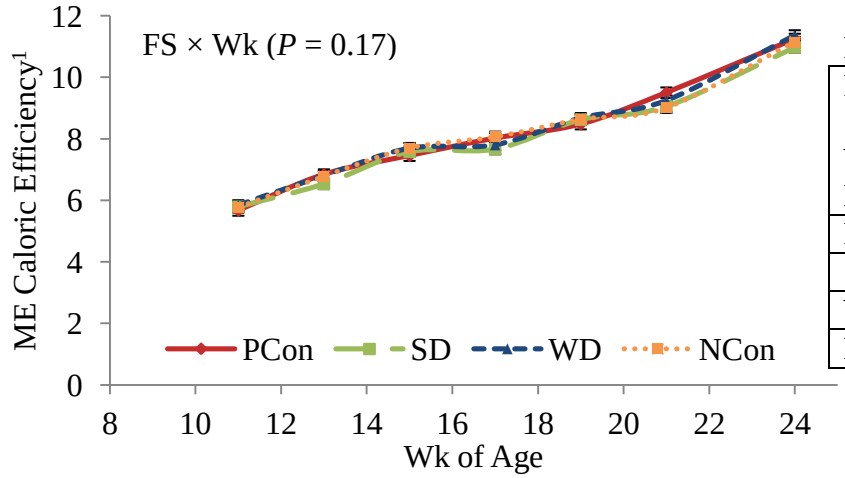
FS[3]	Wk of age 11	13	15	17	19	21	24
PCon							ab
SD							ab
WD							a
NCon							b
PCon	A	B	C	D	DE	E	F
SD	A	B	C	D	D	E	F
WD	A	B	C	D	E	F	G
NCon	A	B	C	D	D	E	F

[1] For each feeding period, SID Lys intake calculated as (total feed intake × calculated dietary SID Lys content)/(pigs per pen × d on feed).
[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.
[3] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a column, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F,G] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 3.7. Calculated ME efficiency of immunologically castrated pigs fed dried distillers grains with solubles feeding strategies (FS) and harvested with increasing time intervals between the second Improvest® dose and harvest (see also Appendix Table A.7 and A.8)



Time interval between 2nd Improvest® dose and harvest[2]	Wk of age						
	11	13	15	17	19	21	24
TD9					a	a	
TD7					b	a	
TD5					ab	b	
TD9	X	AY	B	AB	C	C	D
TD7	A	B	BC	C	C	D	E
TD5	W	XY	AZ	AB	BJ	BH	C



FS[3,4]	Wk of age						
	11	13	15	17	19	21	24
PCon							
SD							
WD							
NCon							
PCon							
SD							
WD							
NCon							

[1] For each period, ME efficiency calculated as (total dietary ME intake/ total BW gain).
[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.
[3] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[4] FS × wk interaction was not significant ($P > 0.05$).
[a,b] Within a column, means without a common superscript differ ($P \leq 0.05$).
[A,B,C] Within a row, means without a common superscript differ ($P \leq 0.05$).
[W,X] Within a row, means without a common superscript differ ($P \leq 0.10$).
[Y,Z] Within a row, means without a common superscript differ ($P \leq 0.10$).
[J,H] Within a row, means without a common superscript differ ($P \leq 0.10$).

Table 3.2. Ultrasound body composition, carcass dressing and fat-free lean percentages, lean gain, and lean gain efficiency of immunologically castrated pigs fed different corn dried distillers grains with solubles (DDGS) feeding strategies (FS) and harvested at 5, 7, or 9 wk post-second Improvest® dose at a commercial abattoir (see also Figure 3.8 and Appendix Table A.11)[1]

	Feeding strategy (FS)[2]					Interval between second Improvest® dose and harvest (TD)[3]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD
Final BW, kg	125.1^{a}	122.7^{ab}	122.4^{ab}	120.3^{b}	1.7	121.6	122.5	123.8	1.7	0.02	0.35	0.96
HCW, kg	90.6^{ax}	88.3^{by}	87.8^{by}	85.3^{b}	1.5	87.8	88.1	88.4	1.5	< 0.01	0.80	0.79
Ultrasound backfat[4], cm	2.09	2.05	2.10	2.02	0.05	2.08^{ab}	2.12^{a}	2.00^{b}	0.05	0.49	0.04	0.03
Carcass dressing[8], %	72.3^{a}	71.8^{ab}	72.0^{ax}	71.1^{by}	0.3	72.0^{ax}	72.0^{a}	71.4^{by}	0.3	0.04	0.04	0.31
Ultrasound LM area[4], cm^2	38.3	37.6	37.2	37.0	1.0	37.4	37.6	37.5	1.0	0.11	0.80	0.96
Lean gain[5], kg/head/d	0.331^{a}	0.328^{a}	0.316^{a}	0.299^{b}	0.013	0.323	0.316	0.318	0.013	< 0.01	0.45	0.40
Lean gain efficiency[6]	0.140^{ax}	0.141^{a}	0.134^{by}	0.130^{b}	0.003	0.135^{a}	0.134^{a}	0.141^{b}	0.003	< 0.01	< 0.01	0.43
Lean gain ME efficiency[7], kg/Mcal	0.0420^{abj}	0.0428^{ax}	0.0408^{bcy}	0.0401^{ck}	0.001	0.0409^{a}	0.0408^{a}	0.0427^{b}	0.001	< 0.01	0.01	0.44
Carcass fat-free lean[9], %	48.7	48.8	48.7	49.1	0.5	48.7	48.9	48.8	0.5	0.35	0.81	0.31

[1] At harvest, one pig was dead in the pen, 15 carcasses required carcass primal trimming. All carcass characteristics for these pigs were removed from the dataset.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.

[3] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at 15, 17, or 19 wk of age corresponding to 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively, before harvest.

[4] HCW included as a covariate ($P \leq 0.001$).

[5] Lean gain per day calculated as (final fat-free lean – initial fat-free lean)/number of d on feed; where final fat-free lean = [(2.620 + (0.401 × HCW, kg) – (3.358 × ultrasound 10th rib backfat depth, cm) + (0.306 × ultrasound 10th rib LM, cm^2) + (0.456 × sex of the pig;barrow=1, gilt=2)] and initial fat-free lean = [(0.922 × initial BW, kg) – 3.65] NPPC (2000).

[6] Lean gain efficiency calculated as total pen fat-free lean gain /total pen feed intake.

[7] Lean gain ME efficiency calculated as total pen fat-free lean gain /total pen metabolizable energy intake.

[8] Carcass dressing percentage was calculated as (final BW before transport/ HCW) × 100.

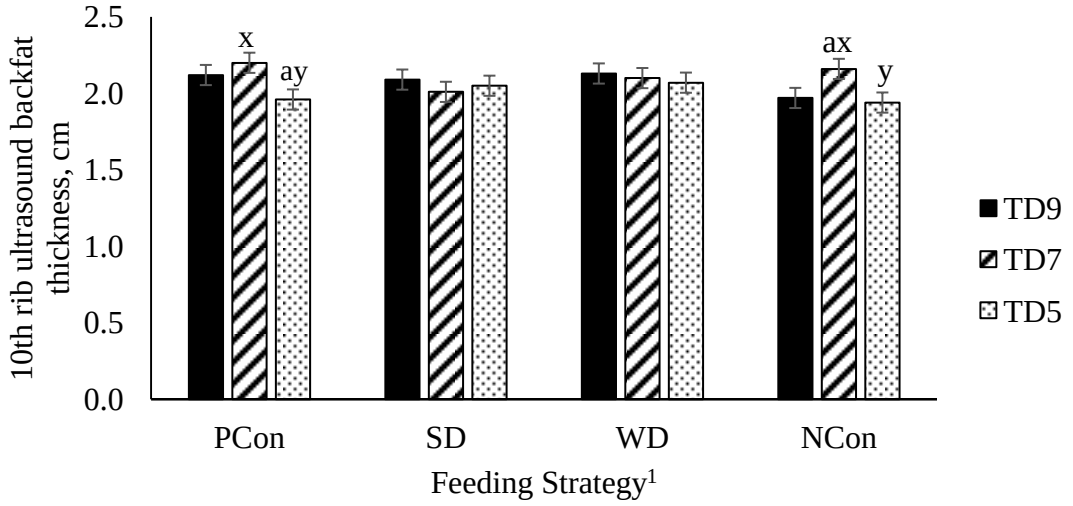
[9] Carcass fat-free lean percentage = [(final fat-free lean)/(HCW)] × 100 (NPPC, 2000).

[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

[j,k] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 3.8. Interaction of using corn dried distillers grains with solubles (DDGS) feeding strategies (FS) and immunologically castrating pigs 5, 7, or 9 weeks before harvest on final 10th rib ultrasound backfat thicknessof pigs harvested at a commercial abattoir (see Appendix Table A.11)[1,2,3]



[1]PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at 15, 17, or 19 wk of age corresponding to 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively, before harvest.

[3] HCW included as a covariate ($P \leq 0.001$).

[a] Means without a common superscript differ ($P > 0.05$).

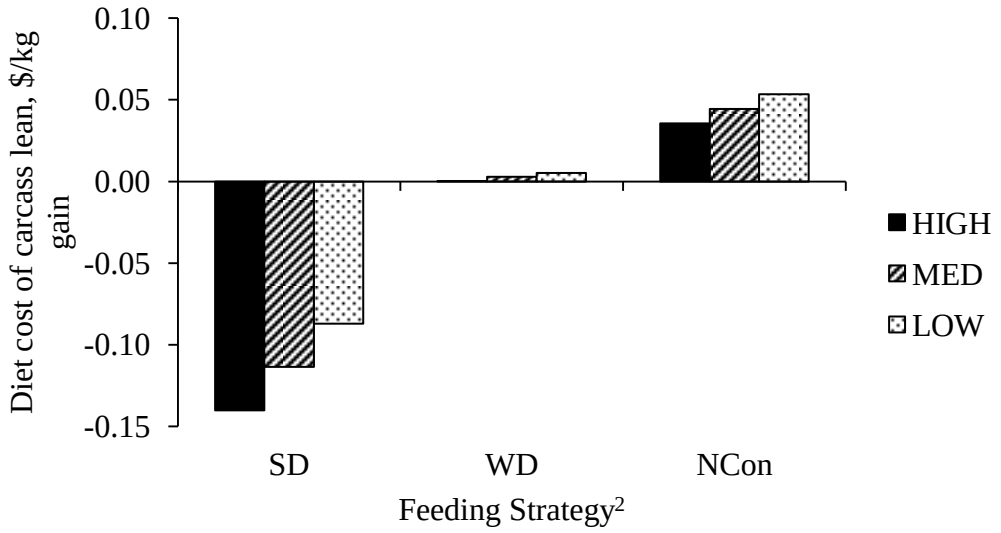
[x,y] Means without a common superscript differ ($P \leq 0.10$).

Table 3.3. Dietary cost of complete feed for each dietary phase fed for each dietary corn dried distillers grains with solubles (DDGS) feeding strategy (FS).

	Dietary Phase[1]			
	1	2	3	4
FS[2]	HIGH ingredient cost,$/kg of complete feed[3]			
PCon	$0.434	$0.413	$0.397	$0.391
SD	$0.400	$0.390	$0.383	$0.382
WD		$0.370	$0.375	$0.391
NCon				$0.370
	MED ingredient cost,$/kg of complete feed[3]			
PCon	$0.366	$0.344	$0.326	$0.319
SD	$0.338	$0.324	$0.315	$0.312
WD		$0.306	$0.311	$0.319
NCon				$0.306
	LOW ingredient cost,$/kg of complete feed[3]			
PCon	$0.298	$0.274	$0.254	$0.247
SD	$0.276	$0.259	$0.247	$0.241
WD		$0.241	$0.247	$0.247
NCon				$0.241

[1] Phase 1 fed for 3 wk, phases 2 and 3 fed for 4 wk each, and phase 4 fed for 5 wk.
[2]PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.
[3] Assumed feed ingredients costs in HIGH, MED, and LOW pricing scenarios: Corn, $0.315, $0.236, $0.157/kg, respectively; Soybean meal, $0.606, $0.551, $0.496/kg, respectively; DDGS, $0.331, $0.276, $0.220/kg, respectively.

Figure 3.9. Reduction of dietary cost of lean gain of corn dried distillers grains with solubles (DDGS) feeding strategies relative to feeding corn-soybean meal diets over three pricing scenarios of immunologically castrated pigs harvested at a commercial abattoir[1]



[1] Assumed feed ingredients costs in HIGH, MED, and LOW pricing scenarios: Corn, $0.315, $0.236, $0.157/ kg, respectively; Soybean meal, $0.606, $0.551, $0.496/kg, respectively; DDGS, $0.331, $0.276, $0.220/kg, respectively.
[2]PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = corn-soybean meal diets containing 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively; WD = corn-soybean meal diets containing 40% DDGS in phases 1 to 3, and 0% DDGS in phase 4; NCon = diets containing 40% DDGS throughout growing-finishing period.

CHAPTER 4: Body composition characteristics of immunologically castrated pigs using different dried distillers grains with solubles feeding strategies determined by ultrasound, serum IGF-1 and leptin during growth, and gene expression of pigs at harvest

I. Summary

Body composition of immunologically castrated (**IC**) pigs was evaluated by live animal ultrasound and serum IGF-1 and leptin concentrations at 3 different time intervals between the second Improvest® dose and harvest, and using 4 different corn dried distillers grains with solubles (**DDGS**) feeding strategies (**FS**). Pigs received the second dose of Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) at 5 (**TD5**), 7 (**TD7**), or 9 (**TD9**) wk before harvest and pigs were fed 1 of 4 DDGS FS in 4 phases for 3, 4, 4, and 5 wk during phase 1 to 4, respectively. Feeding strategies included: 1) corn-soybean meal (**CS**) control diet (**PCon**), 2) CS + 40, 30, 20, and 10% DDGS fed in phase 1 to 4, respectively (**SD**), 3) CS + 40% DDGS fed in phases 1 to 3, and 0% DDGS in phase 4 where pigs were fed CS (**WD**), and 4) CS + 40% DDGS in phases 1 to 4. Pigs (n = 2/pen) were randomly selected at 13 wk of age (**WOA**) for blood collection to determine serum IGF-1 and leptin concentrations by radioimmunoassay. Backfat thickness and LM area were determined by real-time ultrasound, and blood was collected beginning when the second Improvest® dose was administered and continued at 17, 19, 21, and 24 WOA. At 24 WOA, TD7 pigs fed NCon had greater ($P < 0.05$) backfat thickness than TD5 and TD9 pigs fed NCon (2.11 vs. 1.86 and 1.91 ± 0.05 cm, respectively). At 24 WOA, TD7 pigs fed NCon had similar backfat thickness to TD7 pigs fed SD and WD. This is in contrast to TD5 and

TD9 pigs fed NCon, where at 24 WOA, pigs fed PCon, SD, and WD had less ($P < 0.05$) backfat than pigs fed NCon. Prior to removing the DDGS in the WD strategy at 19 WOA, pigs fed WD had similar backfat thickness compared with pigs fed PCon and NCon. At 21 and 24 WOA for TD9 pigs, and at 24 WOA for TD5 pigs fed WD, pigs had greater ($P < 0.05$) backfat thickness compared with pigs fed NCon. Feeding WD also resulted in greater ($P < 0.05$) serum leptin concentration of TD9 pigs compared with TD9 pigs fed NCon at 21 (3.93 vs. 3.02 ± 0.42 ng/mL, respectively) and 24 (4.44 vs. 3.18 ng/mL, respectively) WOA. Among TD treatments, changes serum IGF-1 concentration and LM area were inconsistent in all FS. After removing DDGS from the diet for 5 wk, TD5, TD7, and TD9 pigs fed WD had similar LM area compared to pigs fed PCon. Thus, withdrawing DDGS at 19 WOA resulted in pigs with similar LM area and backfat thickness compared with pigs fed PCon and SD.

KEYWORDS: backfat, dried distillers grains with solubles, feeding strategy, IGF-1, immunologically castrated pigs, leptin

II. Introduction

The time period between the second Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest (**TD**) can range from 3 to 10 wk (FDA, 2011c). As TD increases, ADFI continually increases (Elsbernd et al., 2014). Pigs receiving the second Improvest® dose 9 wk before harvest and fed 40% corn dried distillers grains with solubles (**DDGS**) diets tended to have reduced overall ADFI and ME intake compared with pigs fed corn-soybean meal diets or a feeding strategy (**FS**) where DDGS was removed from the diet 5 wk before harvest (Chapters 2 and 3). This decrease in overall ADFI and ME intake among DDGS

FS was not observed in pigs harvested 5 or 7 wk after the second Improvest® dose (Chapters 2 and 3).

Energy and nutrients are partitioned toward lean deposition, and excess energy is deposited as fat in growing pigs (Noblet and van Milgen, 2013). Regulation of feed intake and the partitioning of nutrients are under hormonal control and partially regulated by energy status and body adiposity (Black et al., 2009). Circulating IGF-1 stimulates protein synthesis and lean growth (Claus and Weiler, 1994). Since IGF-1 declines within 14 d after the second Improvest® dose (Claus et al., 2007), lean growth would also be expected to decline consequently, more nutrients are partitioned toward fat deposition. Moreover, circulating leptin concentration is associated positively with adipocyte mass (Barb et al., 2001). Thus, circulating leptin concentrations should increase after the second Improvest® dose. The objective of this study was to evaluate body composition of IC pigs receiving the second Improvest® dose at 5, 7, or 9 wk before harvest using ultrasound measurements, and determine if serum IGF-1 and leptin concentrations can be used as biological markers for changes in body composition during these time periods. Understanding the real-time changes in body composition after the second Improvest® dose would allow for adjusting diet formulations to match lean and fat deposition requirements and feed intake patterns of IC pigs.

III. Materials and methods

A. Animals and Housing

Intact male pigs (n = 863; GNC V100 Landrace females x V40 Large White boars, Genetiporc, Alexandria, MN) were initially weighed (BW = 21.5 kg) at 8 wk of age (WOA). Pigs were assigned randomly to 1 of 12 treatments in a 4 x 3 factorial

arrangement to include 4 DDGS FS and 3 TD treatments (Chapter 2, Figure 1). Pigs were housed in an environmentally controlled growing-finishing barn with fully slatted floors. Each pen (4.66 x 1.58 m) housed 8 or 9 pigs per pen and contained 1 nipple waterer and 1 four-hole self-feeder. Pigs had *ad libitum* access to feed and water during the 16 wk growing-finishing period. Diets were fed in 4 phases, and each phase was fed for 3, 4, 4, and 5 wk for phases 1 to 4, respectively. Feeding strategies included: positive control (**PCon**) where pigs were fed diets containing 0% DDGS (corn-soybean meal control diet; CS); DDGS step down (**SD**) strategy where pigs were fed CS diets containing 40, 30, 20, or 10% DDGS in phase 1 to 4, respectively; DDGS withdrawal (**WD**) strategy where pigs were fed a CS diet with 40% DDGS in phases 1 to 3 and in phase 4, DDGS was withdrawn from the diet and pigs were fed a CS diet; and negative control (**NCon**) where pigs were fed a CS diet with 40% DDGS throughout the growing-finishing period.

All pigs in this study were immunologically castrated by receiving two, 2-ml s.c. injections of Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) at the post-auricular region of the neck by trained technicians. The first dose was administered to all pigs at 11 WOA. The second dose of Improvest® was administered at 15, 17, or 19 WOA, which corresponded to pigs receiving the second Improvest® dose at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest, respectively. Two wk following the second Improvest® dose, a quality assurance assessment was performed according to Zoetis, Inc. protocol to ensure successful immunological castration. Pigs not meeting specifications (n = 38), received a third dose of Improvest® and remained on the study. At 13 WOA, pigs (n = 2/pen; 192 total) were randomly selected for blood collection throughout the growing-finishing period, and were

harvested at the University of Minnesota Meat Science Laboratory at 24 WOA. The 4 groups of pigs were harvested in July or September of 2012 or January or May of 2013.

B. Diet formulation and dietary composition

Diets were formulated to meet or exceed NRC (2012) requirements for growing IM pigs. All diets were formulated to have a similar SID Lys:ME ratio within a phase. Diet formulations have been described in detail in Chapter 2. Two wk before harvest, group 1 pigs were diagnosed with Hemorrhagic Bowel Syndrome (**HBS**), which was confirmed by necropsy. This prompted the need to orally administer lincomycin (Zoetis, Inc. Florham Park, NJ) until harvest. Group 1 was administered lincomycin through the water for 2 wk before harvest. Group 2 pigs were housed concurrently and were also orally administered lincomycin for 3 wk through the water beginning at 16 WOA. Once the dietary phase changed from phase 3 to 4, diets included 0.044 g/kg diet of lincomycin for 5 wk. Due to the known health history of this source of pigs, group 3 pigs were administered 0.044 g/kg diet of lincomycin throughout the entire growing-finishing period. However, 10 d before harvest of group 3 pigs, another mortality occurred due to HBS, resulting in an increased dosage of feed-administered lincomycin to 0.088 g/kg diet. Group 4 pigs were fed 0.088g/kg diet of lincomycin throughout the growing-finishing period with the addition of water-medicated Denagard® for 6 d at 10 WOA to treat Illeitis.

C. Body composition and serum leptin and IGF-1

Ultrasound evaluation and blood collection were initiated when the second Improvest® dose was administered, and continued at 17, 19, 21, and 24 WOA. Measurements using B-mode ultrasonography (Aloka 500V SSD; Hitachi-Aloka Medical

Ltd., Tokyo, Japan) were collected for 10th rib backfat and LM area for all pigs and performed by a single trained and certified technician. Pigs were restrained by nose snare and 10 mL of blood was collected via venipuncture of the jugular vein using an 18 gauge x 3.81 cm needle into glass vacutainer tubes. Blood was immediately placed on ice and refrigerated at 4°C overnight for maximum serum harvest. The following morning, the centrifuge was pre-cooled and blood was centrifuged at 1100 × g for 20 min at 4°C. Serum was aliquoted into labeled microcentrifuge tubes and stored at -20°C for later determinations of leptin and IGF-1 concentrations by radioimmunoassay (**RIA**). Before RIA, serum was pooled by experimental pen. Serum leptin concentrations were determined using 100 µl of serum with a commercially available multi-species RIA kit (#XL-85K Millipore, St. Charles, MO) following the manufacturer's instructions. Modifications to the manufacturer's protocol included performing assays in triplicate, and adding an additional standard curve point of 1.04 ng/ml human equivalents (**HE**). Assay tubes were aspirated and radioactivity of the remaining pellet was determined by gamma counter (Packard COBRA II Gamma Counter, Packard Biosciences, Boston, MA). The minimum sensitivity level of this assay was specified by the manufacturer to be 1.00 ng/mL human equivalents. Serum concentrations of IGF-1 were assayed in triplicate as previously described by Lamberson et al., 1995). Intraassay CV for the IGF-1 assay was 2.1%.

D. Pig harvest and tissue collection

Pigs were transported 260 km on the day before harvest, and had *ad libitum* access to water only after transportation. The morning of harvest, pigs were transported 0.7 km to the University of Minnesota Meat Science Laboratory. Pigs were harvested by

electrical stunning and exsanguinated. Carcasses were dehaired by scalding. Carcasses were eviscerated and split down the mid-line. The semimembranous muscle (**SM**) and s.c. last rib backfat were excised at a mean of 20.9 ± 3.9 min after stunning. Each tissue from each animal was collected using an aseptic scalpel blade and autoclaved disposable forceps. Excised tissues were placed in a clean, unused weigh boat, and tissues were cut into pieces less than 0.5 cm thick, as recommended by the manufacturer of the RNA stabilization reagent. Backfat (300 mg) and SM (100 mg) tissues were submerged into 5.0 and 2.0 ml tubes, respectively, that contained 3 mL and 1 mL of RNAlater (Qiagen #76106, Valencia, CA). Prepared tissue samples were immediately placed on ice. Following harvest of each group of pigs, tissue samples were confirmed to be submerged in RNAlater and stored at 4°C. Extraction of RNA was completed 2 to 4 wk after tissue collection. After RNA extraction, RNAlater was decanted from each sample and samples were frozen at -80°C for archival storage.

E. RNA extraction and cDNA synthesis

The RNA was extracted from each sample using RNeasy Mini Kit (Qiagen #74104, Valencia, CA). Concentrations of RNA were determined by spectrophotometric absorbance at 260 nm. Each sample of RNA was portioned into one, 2-µg aliquot, and the remaining RNA was portioned into 1-µg aliquots. Aliquots were frozen at -80°C. The 2-µg aliquot was used to confirm RNA quality by agarose-formaldehyde gel electrophoresis and ethidium bromide staining. A 1-µg aliquot of RNA was reverse-transcribed using Tacq-Man Reverse Transcriptase Reagents (Applied Biosystems, Foster City, CA) to synthesize cDNA as recommended by the manufacturer. A pool of cDNA was

synthesized to serve as a control and was included on each PCR plate. The cDNA was stored at 4°C until PCR analysis.

F. Real-time RT-PCR

Semimembranosus muscle and s.c. backfat mRNA of leptin and IGF-1 were amplified using real-time RT-PCR. The relative quantity of leptin and IGF-1 was measured in quadruplicates by SYBR Green PCR Master Mix (Applied Biosystems, Foster City, CA). The SYBR Green Master Mix was combined with 1µl of cDNA, and leptin or IGF-1 forward and reverse primers (Table 4.1) for PCR amplification. The PCR amplification and analysis was conducted by GeneAmp 5700 Sequence Detection System (Applied Biosystems, Foster City, CA) using 40 cycles of 15 s at 95°C and 1 min at 60°C. Expression of leptin and IGF-1 mRNA was calculated as a proportion to cyclophilin expression for each sample.

G. Statistical analysis

Ultrasound backfat and LM area, as well as serum leptin and IGF-1 data were analyzed using PROC GLMMIX in SAS (Cary, NC). The model statement included FS, TD, time, FS × time, TD × time, time × time, FS × TD × time, and time × time × FS × TD interactions. The time × time polynomial tests for a curvilinear response of the variables over time. Pig group was included as a random effect. Least square mean estimates were separated and adjusted using the Bonferroni option. The mRNA gene expression of adipose and muscle leptin and IGF-1 concentrations were analyzed using PROC MIXED in SAS (Cary, NC). The model included FS, TD, and the FS × TD interaction. Least square means were separated and adjusted using the Tukey option. Pig group was included as a random effect and pen was considered the experimental unit. For

each variable, the residuals were used to examine the assumptions of normality using PROC Univariate. Significance was determined when $P \leq 0.05$ and trends were identified when $P \geq 0.05$ and ≤ 0.10.

IV. Results and discussion

Lean and adipose tissues represent more than 80% of carcass components in market weight pigs (Lawrence et al., 2012). Lean tissue is more efficiently deposited than adipose tissue (Noblet and van Milgen, 2013) and has the greatest product value for human consumption (Lawrence et al., 2012). Thus, the partitioning of energy and nutrients toward lean and adipose accretion is of economic importance. After the second Improvest® dose, IC pigs transition toward growth patterns more like physical castrates (**PC**), where feed intake of IC pigs rapidly and continually increases as the time interval between the second dose and harvest is extended (Lealiifano et al., 2011, Elsbernd et al., 2014). This increase in feed intake subsequently leads to increased intake of energy and nutrients (Chapter 3). These changes in feed intake occur during a time when lean accretion typically declines and adipose accretion increases.

A. Effect of timing of the second Improvest® dose and DDGS feeding strategy on LM area after the second Improvest® dose

In IC pigs, Tavarez et al. (2014) reported that LM area (determined by live animal ultrasound) increased when increasing the interval between the second Improvest® dose and harvest, and was greater in IC pigs compared with PC harvested 8 wk after the second Improvest® dose. In that study, Tavarez et al. (2014) administered the second Improvest® dose to all pigs of similar age, but increased the time interval between the second Improvest® dose and harvest by increasing pig age. Therefore, it is difficult to

distinguish if the increase in LM area was due to increasing age of IC pigs or due to increasing the time interval between the second Improvest® dose and harvest. In contrast, the present study, evaluated LM area among pigs of the same chronological age, but where the second Improvest® dose was administered to pigs at 15, 17, or 19 WOA.

In the current study, since energy is partitioned toward lean and fat, the reduced ME intake/d of TD9 pigs fed NCon compared with TD9 pigs fed PCon and WD (in Chapter 3) was expected to reduce lean accretion in TD9 pigs fed NCon. However, this did not occur because the LM area was not different among TD treatments within each FS (Figure 4.1 and Appendix Table A.12). For each TD treatment, lean growth was not improved in pigs fed NCon as observed by reduced ($P < 0.05$) LM area compared with pigs fed PCon. This occurred from 17 to 24 WOA in TD7 and TD9 pigs and at 19 to 24 WOA in TD5 pigs (Figure 4.2 and Appendix Table A.12). Only 1 other study has evaluated feeding DDGS to IC pigs and the use of a DDGS withdrawal feeding strategy, but LM area and depth was determined only in carcasses (Asmus et al., 2014). In the Asmus et al. (2014) study, no difference in carcass LM area and depth was observed when feeding corn-soybean meal, corn-soybean meal with 30% DDGS, or when removing DDGS from the diet for 5 or 7 wk before harvest in either IC or PC pigs.

Increasing dietary inclusion rates of DDGS can have a greater adverse effect on lean growth. Relative to gilts and PC fed PCon, pigs fed diets containing 20% DDGS had similar LM area (Hilbrands et al., 2013), but feeding diets containing 30 (Song, 2013), 40 (Hardman, 2014), or 60% (Hardman, 2014) DDGS resulted in reduced LM area or depth at harvest. Hilbrands et al. (2013) determined that the negative effect of feeding 40% DDGS on LM area, relative to feeding corn-soybean meal diets, is more apparent when

feeding low digestible AA DDGS sources. Feeding 40% high AA digestibility DDGS sources restored LM area, such that LM area was similar to pigs fed corn-soybean meal diets. Thus, differences in AA digestibility among DDGS sources (Urriola et al., 2013b) is of greater concern when adding it to diets at high inclusion rates for suboptimal lean growth because of the potential to overestimate AA digestibility of the DDGS source fed.

At 24 WOA in the present study, SD and WD were effective feeding strategies in overcoming the reduced ($P < 0.05$) LM area of TD5, TD7, and TD9 pigs fed NCon (Figure 4.2 and Appendix Table A.12). Pigs fed WD had reduced ($P < 0.05$) LM area compared to pigs fed PCon, but only until 21 WOA. When DDGS was removed from the diet 5 wk before harvest, pigs fed WD had similar LM area compared with pigs fed PCon at 24 WOA. Pigs fed SD had reduced ($P < 0.05$) LM area compared with pigs fed PCon until 19 WOA. At 21 and 24 WOA, LM area was similar in TD5, TD7, and TD9 pigs fed SD and PCon. The increased lean growth in the final dietary phase was likely the result of greater ADFI of a diet that had greater digestibility of AA and higher ME density. Thus, higher levels of DDGS (at least 40%) can be included in growing-finishing swine diets without compromising lean growth if highly digestible AA DDGS sources are used or DDGS is removed from the diet at least 5 wk or gradually removed before harvest.

B. Effect of timing of the second Improvest® dose and DDGS feeding strategy on backfat thickness after the second Improvest® dose

Most studies have measured body composition in IC pigs in the carcass at harvest. Several researchers have reported that backfat depth of IC pigs is reduced (Dunshea et al., 2001; Pauly et al., 2009; Boler et al., 2012; Yuan et al., 2012), or tends (Asmus et al., 2014b) to be reduced, compared with PC. In the present study, live animal

ultrasound evaluation was used to determine changes in body composition after the second Improvest® dose was administered. For each FS, backfat thickness was not different between TD9 and TD7 pigs at 17 WOA (Figure 4.3 and Appendix Table A.12). This is interesting because TD9 pigs received the second Improvest® dose 2 wk earlier and thus, it was expected that backfat thickness would have been greater in TD9 than TD7 pigs. However, as expected, at 19 WOA, pigs receiving the second Improvest® dose (e.g. TD5 pigs) had less ($P < 0.05$) backfat than TD9 pigs when fed PCon (1.36 vs. 1.49 ± 0.05 cm, respectively), SD (1.43 vs 1.32 ± 0.05 cm, respectively), or WD (1.41 vs. 1.30 ± 0.05 cm, respectively) feeding strategies, and TD5 pigs also tended ($P < 0.10$) to have less backfat than TD9 pigs when fed NCon (1.24 vs 1.34 ± 0.05 cm, respectively). When beginning ultrasound evaluation at 22 WOA, Travarez et al. (2014) determined that increasing the time interval between the second Improvest® dose and harvest increased backfat thickness of PC and IC pigs at 22 to 28 WOA (2 to 8 wk after the second Improvest® dose). However, the increase in backfat thickness at 8 wk after the second Improvest® dose was greater in PC than IC pigs (Tavárez et al., 2014b).

In the present study, pigs were harvested at 24 WOA. At that time, backfat thickness was similar among TD treatments within WD and SD (Figure 4.3 and Appendix Table A.12). In contrast, at 24 WOA, TD7 pigs fed NCon had increased ($P < 0.05$) backfat thickness compared with TD9 and TD5 pigs (2.11 vs. 1.91 and 1.86 ± 0.05 cm, respectively). The difference in backfat thickness among TD treatments within the NCon feeding strategy is due to TD7 pigs fed NCon having similar backfat thickness to pigs fed SD and WD (Figure 4.4 and Appendix Table A.12). This did not occur in TD5 and TD9 pigs. In contrast, TD5 and TD9 pigs fed NCon had less ($P < 0.05$) backfat than

pigs fed SD, and WD at 24 WOA (TD9: 1.91 vs. 2.14 and 2.17 ± 0.05 cm, respectively; TD5: 1.86 vs. 2.05 and 2.05 ± 0.05 cm, respectively; Figure 4.4 and Appendix Table A.12). This discrepancy in backfat thickness at 24 WOA in pigs fed NCon can be explained by decreased overall ME intake/d among TD9 pigs and similar overall ME intake/d among TD7 pigs (Chapter 3). However, overall ME intake/d of TD5 pigs was also similar among FS, which does not support the reduced backfat thickness of TD5 pigs fed NCon compared with TD5 pigs fed PCon, SD, or WD. Asmus et al. (2014) observed that backfat depth was not different among pigs fed a corn-soybean meal diet, a corn-soybean meal plus 30% DDGS diet, and corn-soybean meal diets with the 30% DDGS removed 5 or 7 wk before harvest. Additionally, the dietary effect on backfat depth was similar in IC and PC pigs (Asmus et al., 2014b). When feeding 40% DDGS to gilts and PC pigs, Hilbrands et al. (2013) used live animal ultrasound to show that feeding diets containing 40% of a highly digestible AA DDGS source resulted in less backfat compared with pigs fed corn-soybean meal diets, which was likely due to the use of AA and energy for greater lean deposition. In the present study, overall SID Lys intake/d followed a similar pattern to overall ME intake/d. While LM area was reduced in all TD treatments fed NCon strategy compared with pigs fed PCon, backfat thickness appeared to be affected to a greater extent suggesting that energy intake was the limiting factor in altering body composition.

C. Physiological responses to changes in body composition

Voluntary feed intake and lean and adipose growth are hormonally regulated (Black et al., 2009). Intact male pigs have greater circulating concentrations of estradiol and testosterone compared with PC and gilts (Clapper et al., 2000). These hormones

stimulate protein synthesis by increasing growth hormone and IGF-1 secretion (Claus et al., 1994a), and thus, regulate the partitioning of energy and nutrients toward lean growth and away from fat deposition. In this study, all TD5 pigs, regardless of FS treatment, had greater serum IGF-1 concentrations compared with TD9 pigs at 19 WOA (Figure 4.5 and Appendix Table A.13). Given that serum estradiol-17β and IGF-1 concentrations increase over time in IM pigs until about 126 (Clapper et al., 2000) or 154 d of age (Batorek et al., 2012), delaying the administration of the second Improvest dose may allow capturing more of the endogenous anabolic growth potential of IC pigs. Typically, within the first 14 d following the second Improvest® dose, circulating concentrations of LH, testosterone, estradiol 17-β (Claus et al., 2008), and IGF-1 concentrations decrease, while blood urea nitrogen concentration increases (Claus et al., 2008; Huber, 2012), indicating that protein synthesis and lean growth are reduced. Since TD9 pigs received the second Improvest dose 4 wk before TD5 pigs, it was expected that IGF-1 would be reduced at 19 WOA in TD9 pigs compared with TD5 pigs. Additionally, at 19 WOA, TD5 pigs fed WD and NCon also had greater ($P < 0.05$) serum IGF-1 concentrations compared with TD7 pigs (Figure 4.5 and Appendix Table A.13). Up to 19 WOA, pigs fed WD and NCon were consuming the same diets containing 40% DDGS. The differences in serum IGF-1 concentration were not consistent with live animal ultrasound for LM area due to the lack of differences observed in LM area between WD and NCon at 19 WOA.

Among TD9 pigs, pigs fed SD had greater ($P < 0.05$) serum IGF-1 concentrations than pigs fed PCon at 15 (214.5 vs. 169.0 ± 19.3 ng/mL, respectively) and 17 WOA (206.7 vs. 166.7 ± 19.3 ng/mL, respectively; Figure 4.6 and Appendix Table A.13). This was also observed in TD7 pigs where pigs fed SD had greater ($P < 0.05$) serum

concentrations of IGF-1 at 17 WOA (232.2 vs. 193.0 ± 19.3 ng/mL, respectively) compared with pigs fed PCon. As the growing-finishing period progressed, the differences in serum IGF-1 concentrations among FS diminished. In TD7 and TD9 pigs, serum IGF-1 concentrations among FS were similar at 19, 21, and 24 WOA. However, in TD5 pigs at 19 WOA, pigs fed SD tended ($P < 0.10$) to have greater serum IGF-1 concentrations than pigs fed PCon (231.2 vs. 200.4 ± 19.3 ng/mL, respectively). Like TD7 and TD9 pigs, the effect of FS on serum IGF-1 concentrations in TD5 diminished, and serum IGF-1 concentrations were similar at 21 and 24 WOA. These results contradict the expected lower serum IGF-1 concentrations of pigs fed NCon because the LM area of TD5 pigs fed NCon was less than in pigs from other FS. This suggests that serum IGF-1 concentrations should have been reduced in pigs fed NCon. In addition, considering the increase in ADFI, overall ME intake/d, and LM area of pigs fed WD, serum IGF-1 concentrations should have also increased.

The discrepancy between LM area and serum IGF-1 concentration could have been confounded by the health challenges of HBS. The diagnosis of HBS prompted the need to orally-administer Lincomycin as a therapeutic treatment. As a result, pigs in groups 3 and 4 were also placed on antibiotics as a preventative measure before clinical signs or mortalities occurred. Use of feed antibiotics (ASP-250) has been shown to increase serum IGF-1 concentrations (Hathaway et al., 1996). However, in this study Lincomycin was administered and it is not known if Lincomycin has the same effect on increasing serum concentration of IGF-1 as ASP-250. Hemorrhagic bowel syndrome is an enteric condition that can result from pathogenic causes due to *Lawsonia intracellularis* infection, or animal behaviors such as increased activity, fighting, or

irregular feeding (Harris, 2013). In this study, evaluation and recording of behavioral changes of pigs was not an experimental objective. However, other researchers have observed that at 17 WOA (1 wk before the second Improvest® dose was administered) pigs designated for immunological castration had similar social behaviors to IM pigs, including aggressive and mounting behaviors (Cronin et al., 2003). After the second Improvest® dose, IC pigs had similar behavior to PC (Cronin et al., 2003). However, in this study, mortalities due to HBS occurred after pigs received the second Improvest® dose. As a result, the occurrence of HBS may not have been behaviorally related.

Leptin is an anorexigenic hormone secreted by adipocytes. Circulating leptin concentrations are directly proportional to body adipose mass, and thus, leptin serves as an energy sensor (Barb et al., 2001). Since circulating anabolic hormones promote greater lean deposition and minimize fat deposition (Claus and Weiler, 1994), IM pigs are expected to have reduced circulating leptin concentrations compared with IC and PC pigs. In the current study, when pigs were fed PCon and NCon, TD5 pigs had lower ($P < 0.05$) serum leptin concentrations than TD9 pigs at 19 WOA (PCon: 2.36 vs. 3.36 ± 0.42 ng/ml; NCon: 1.85 vs. 2.75 ± 0.42 ng/ml; Figure 4.7 and Appendix Table A.13). However, when pigs were fed SD or WD feeding strategies, serum leptin concentrations were similar among TD treatments. Our current understanding is that when body fat mass increases, circulating leptin increases and results in reduced feed intake (Barb et al., 2001). However, Batorek et al. (2012) reported that IM and PC pigs have similar serum leptin at 83 and 130 d of age despite IM pigs having reduced ADFI compared with PC pigs. While serum leptin concentrations did not increase in IM pigs from 130 to 154 d of age, serum leptin of IC and PC pigs increased over the same time period (Batorek et al.,

2012). While changes in backfat and feed intake were not evaluated over time by Batorek et al. (2012), IM pigs had less ADFI, backfat, and serum leptin compared with PC at the end of the study. Therefore, the assumption that increasing backfat results in increasing serum leptin concentration, and thus reduced feed intake, may not occur in IM pigs. Intact male pigs produce large quantities of estradiol-17 β, and in fact, can have more than 5 times greater circulating estradiol-17 β concentrations than gilts (Clapper et al., 2000). Estradiol-17 β can decrease feed intake (Wade and Zucker, 1970) and may be a factor in modulating the relationship between feed intake, serum leptin concentrations, and backfat. This may explain the lack of a consistent relationship among backfat thickness, feed intake, and serum leptin concentrations observed in this study and the variable differences in serum leptin concentration of TD5 and TD9 pigs among FS.

D. mRNA expression of IGF-1 and leptin in muscle and adipose tissues

In general, circulating IGF-1 is less important than locally produced IGF-1 in regulating muscle growth (Hossner, 2005). Thus, evaluation of mRNA expression for IGF-1 and leptin in muscle and adipose tissues was conducted. The differences in LM area among FS suggest that mRNA expression of IGF-1 would be greater in pigs fed PCon than those fed NCon. However, adipose and muscle IGF-1 (Figure 4.8) and leptin (Figure 4.7) mRNA expression were not different among TD treatments and FS in this study. It is not clear why differences were observed in LM area and backfat thickness at 24 WOA, but not in IGF-1 and leptin mRNA expression. It is possible that no differences in mRNA expression were detected due to withholding pigs from feed for approximately 18 h before harvest. Previous studies have evaluated “acute” 72 h feed withdrawal and have shown that leptin and IGF-1 mRNA expression in adipose tissue was lower when

feed was withheld for 72 h compared with pigs provided *ad libitum* to feed (Salfen et al., 2003). Additionally, after a 24 h re-alimentation period, mRNA expression of pigs withheld from feed remained lower than for pigs provided *ad libitum* access to feed (Salfen et al., 2003). However, these researchers did not evaluate mRNA expression in muscle. In pigs, data are limited regarding the effect of shorter feed withdrawal periods on mRNA expression. Therefore, it is difficult to ignore possible effects that feed withdrawal of 18 h may have had on mRNA expression in this study.

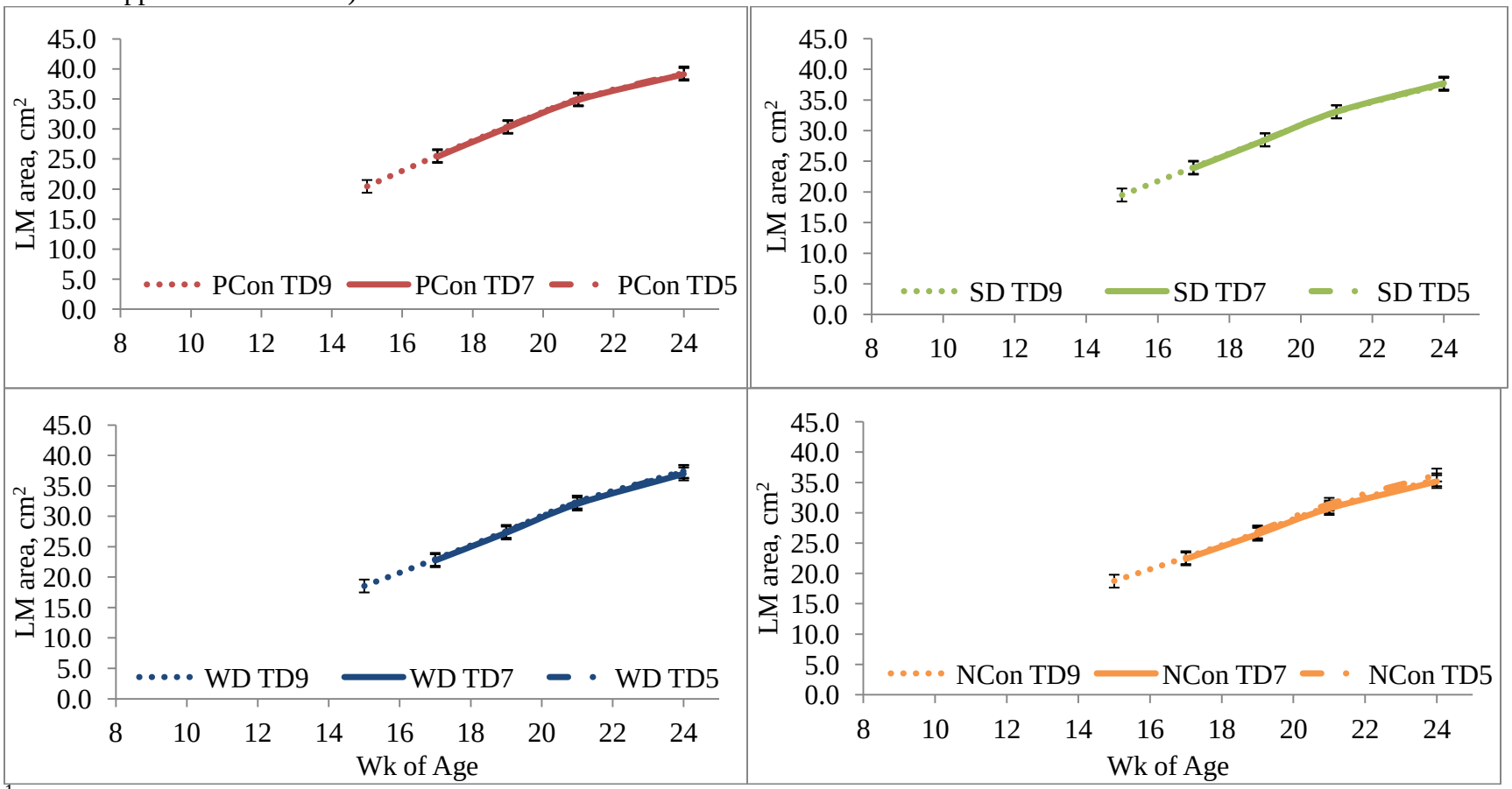
In conclusion, pigs fed NCon had reduced LM area in all TD treatments compared with pigs fed PCon beginning at 17 WOA. Pigs fed NCon also had reduced backfat thickness in TD9 beginning at 17 WOA, and in TD5 pigs beginning at 19 WOA. These differences in LM area and backfat thickness among pigs fed NCon and PCon persisted until 24 WOA. These responses were likely due to a similar reduction in overall ME intake/d between pigs fed NCon and PCon (Chapter 3, Table 3.1). Additionally, pigs fed WD had less backfat than pigs fed PCon from 17 to 21 WOA in all TD treatments. However, at 24 WOA, LM area was similar between pigs fed PCon and WD in all TD treatments. For TD5 and TD9 pigs fed WD, backfat thickness also increased once the DDGS was removed from the diet so that backfat thickness was similar with pigs fed PCon and SD at 24 WOA. This indicates that for TD5 and TD9 pigs, increased ME intake due to removing the DDGS was partitioned to both lean and fat accretion. However, this did not occur in TD7 pigs because pigs fed NCon and WD had similar backfat thickness at 24 WOA due to similar ME intake/d. It is unclear why TD7 pigs had a different response to ME intake and nutrient partitioning compared with TD5 and TD9 pigs. These results suggest that there are complex interactions that regulate energy intake

and nutrient partitioning in IC pigs. Therefore, further investigations are needed to determine the dietary limitations of feed intake related to the changes in endocrine profile and age when the second Improvest® dose is administered.

Table 4.1. Forward and reverse primers for PCR of IGF-1 and leptin mRNA

Primer	Sequence
IGF-1	
Forward	5’ GCT CGT GGA CGC TCT TCA GT 3’
Reverse	5’ ATC CAC GAT GCC CGT CTG T 3’
Leptin	
Forward	5’ GAC CCC TGT GCC GAT TCC 3’
Reverse	5’ GAC AGA CTG CAT GTG TGA AAT GTC 5’
Cyclophilin	
Forward	5’ GGT CCT GGC ATC TTG TCC AT 3’
Reverse	5’ TGG CAG TGC AAA TGA AAA ACT G 3’

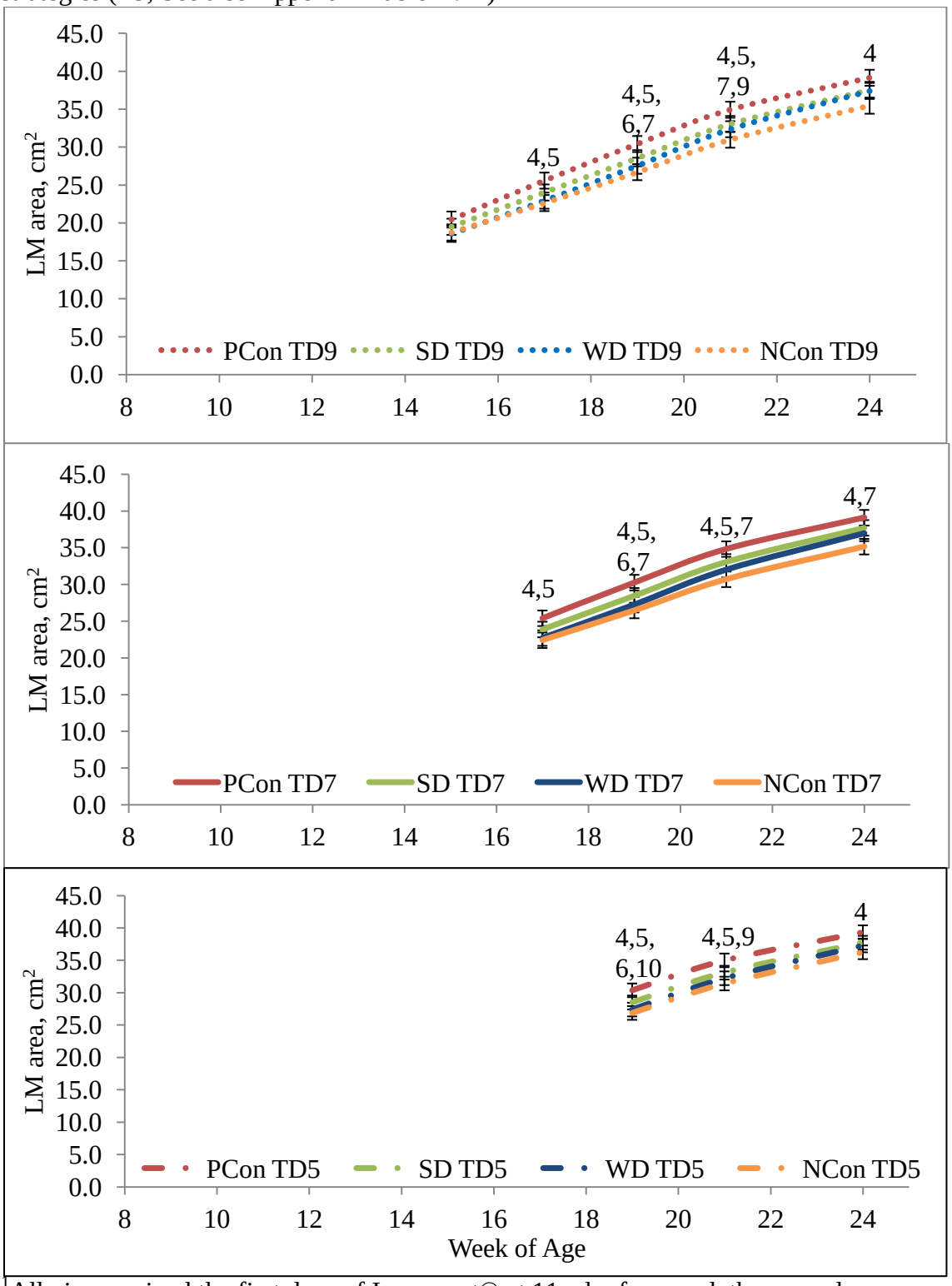
Figure 4.1. Longissimus muscle (LM) area after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.12)[1,2,3]



[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 (P = 0.46).

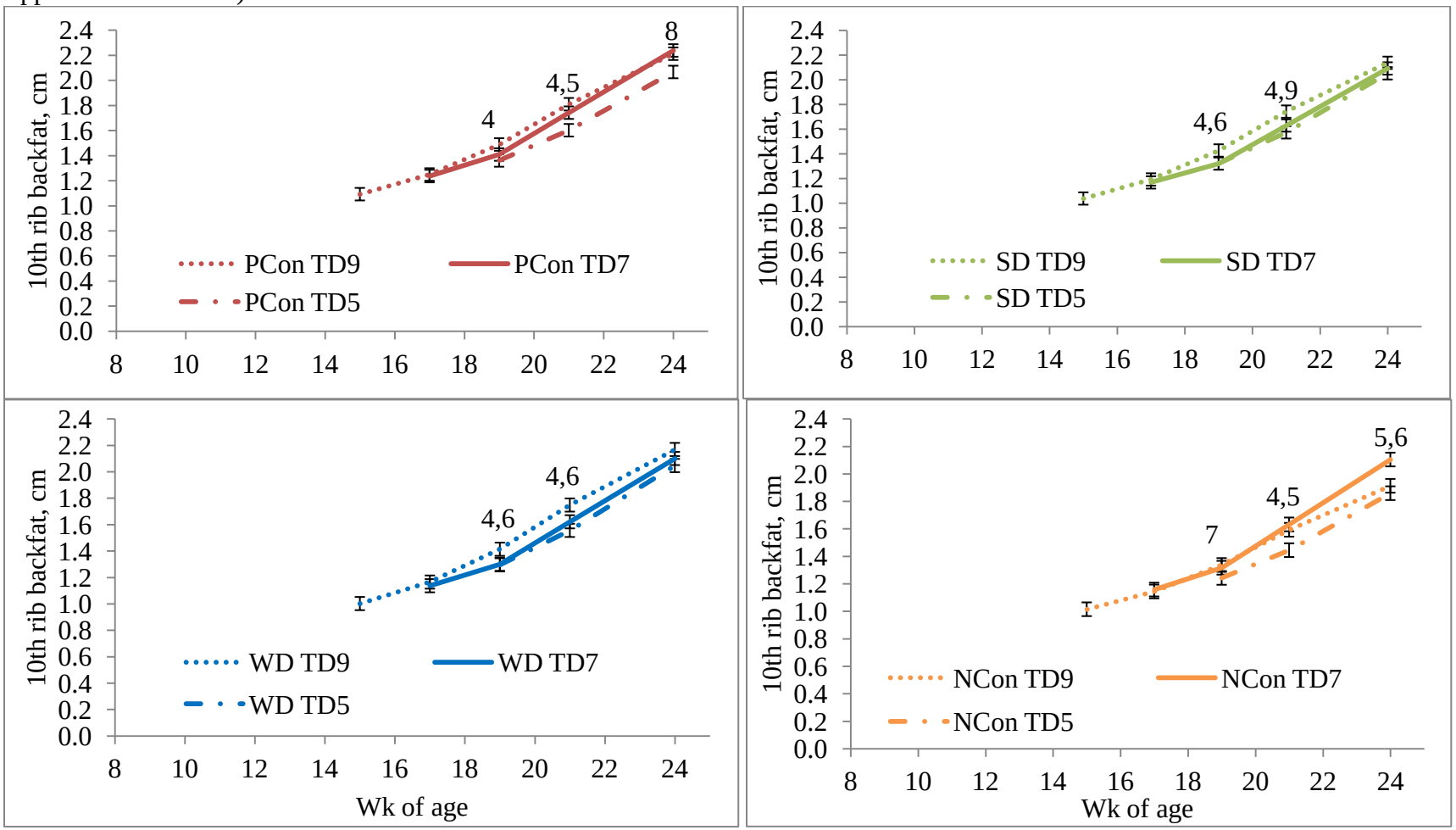
Figure 4.2. Longissimus muscle (LM) area after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.12)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second

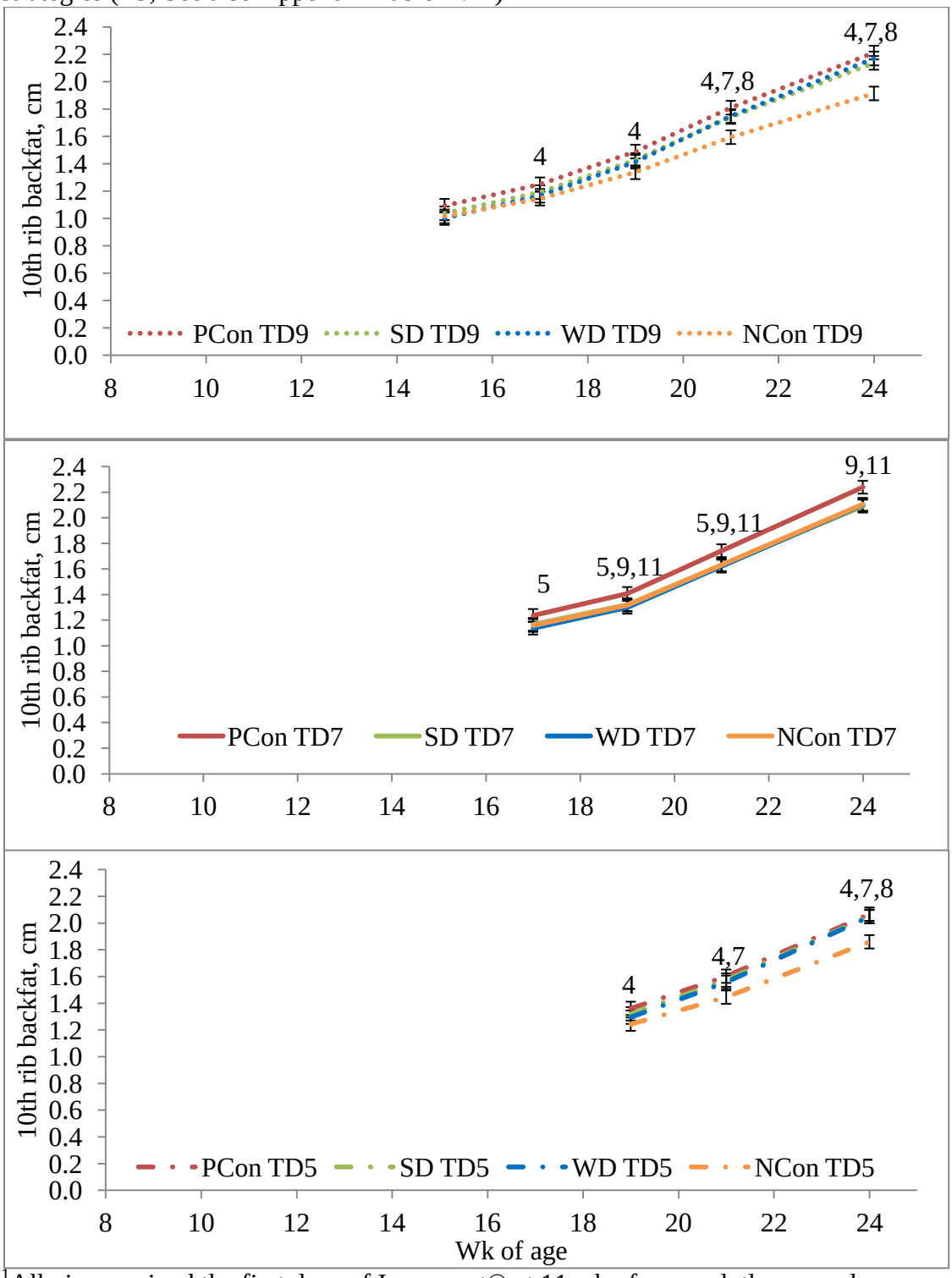
Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P < 0.46$).
[4] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.05$).
[5] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.05$).
[6] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.05$).
[7] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.05$).
[8] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.05$).
[9] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.10$).
[10] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.10$).
[11] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.10$).
[12] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.10$).
[13] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.10$).

Figure 4.3. Backfat deposition after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.12)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P < 0.05$).
[4] Within in each DDGS feeding strategy at each time period, TD5 vs. TD9 ($P < 0.05$).
[5] Within in each DDGS feeding strategy at each time period, TD5 vs. TD7 ($P < 0.05$).
[6] Within in each DDGS feeding strategy at each time period, TD7 vs. TD9 ($P < 0.05$).
[7] Within in each DDGS feeding strategy at each time period, TD5 vs. TD9 ($P < 0.10$).
[8] Within in each DDGS feeding strategy at each time period, TD5 vs. TD7 ($P < 0.10$).
[9] Within in each DDGS feeding strategy at each time period, TD7 vs. TD9 ($P < 0.10$).

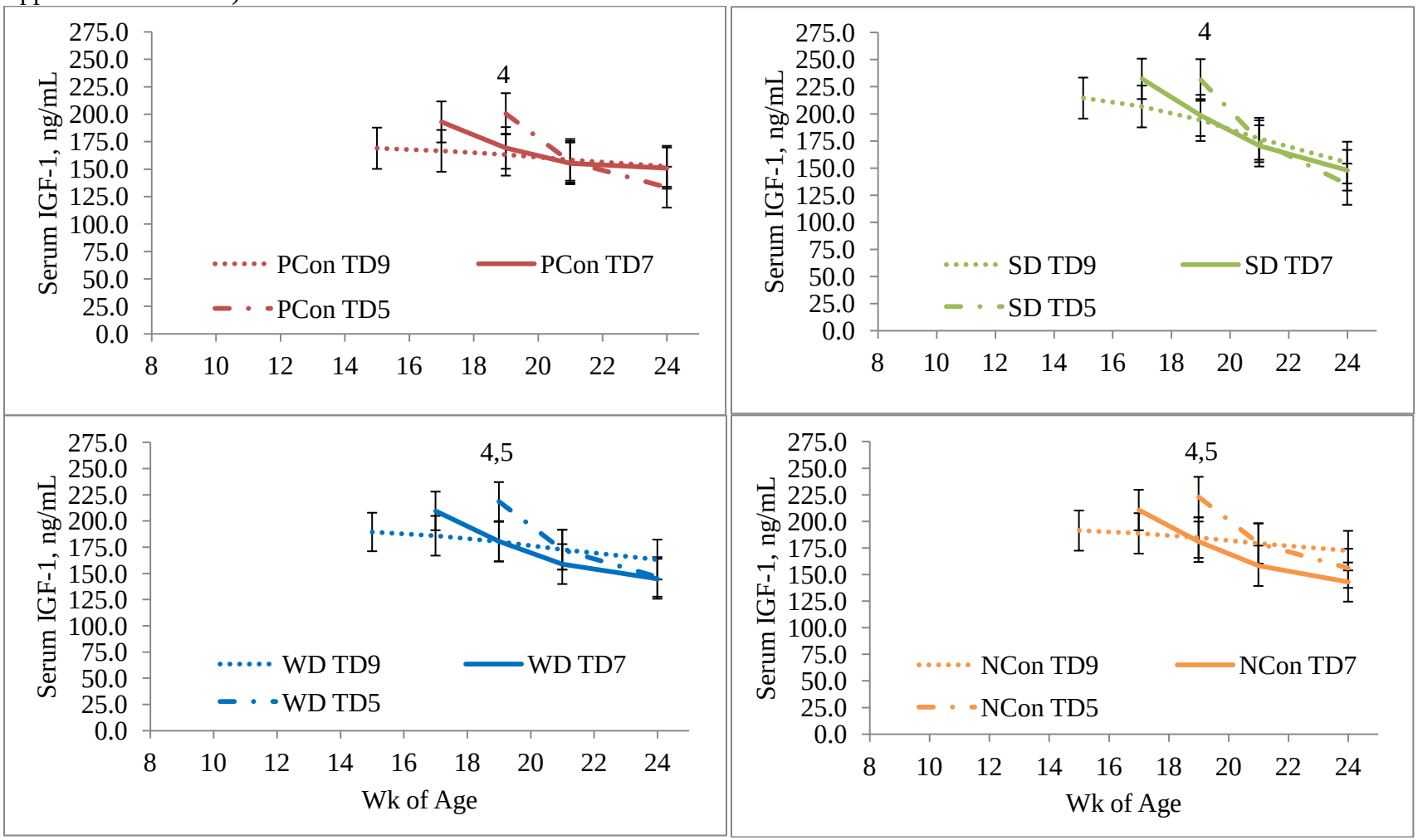
Figure 4.4. Backfat deposition after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.12)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded

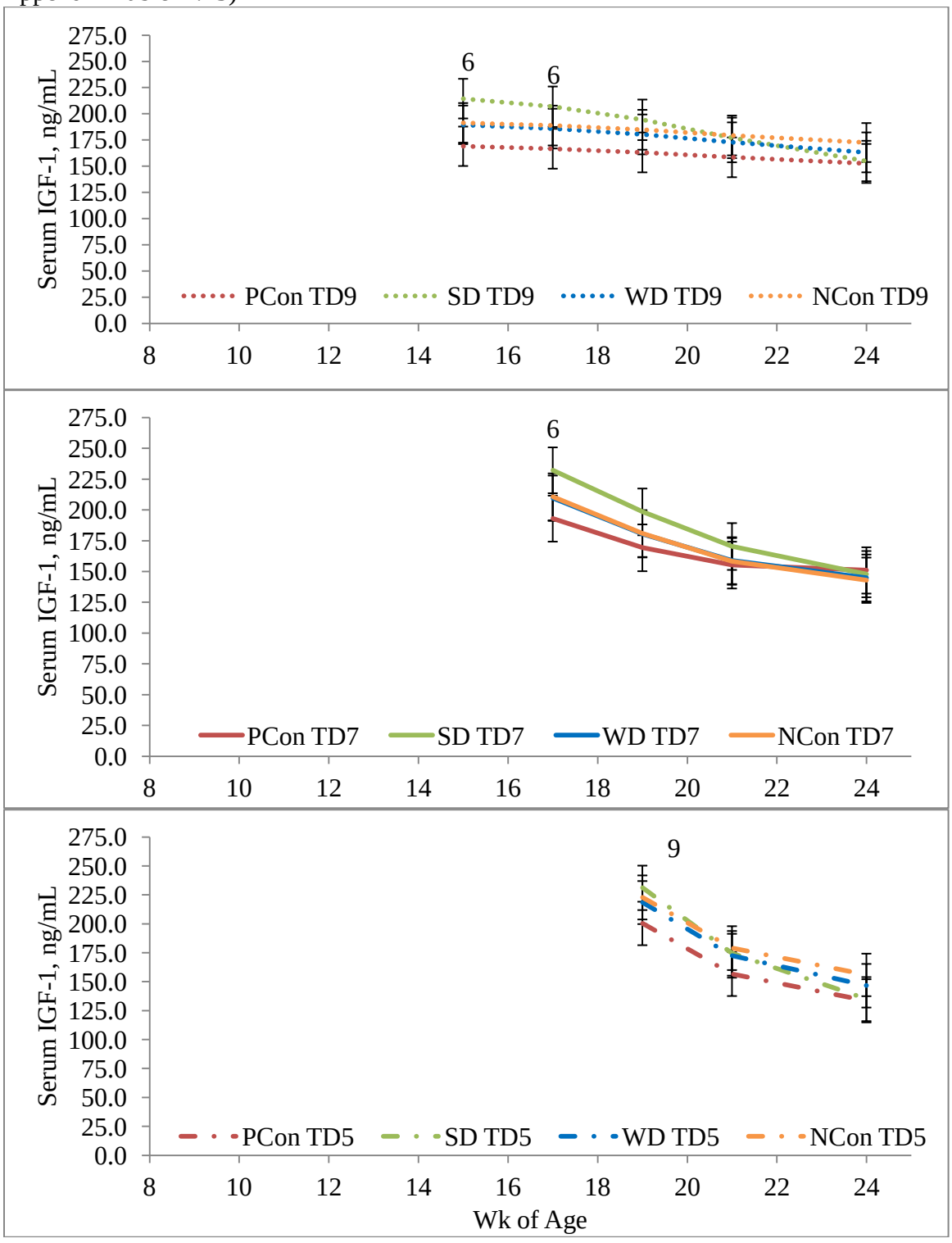
to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P < 0.05$).
[4] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.05$).
[5] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.05$).
[6] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.05$).
[7] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.05$).
[8] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.05$).
[9] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.10$).
[10] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.10$).
[11] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.10$).
[12] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.10$).
[13] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.10$).

Figure 4.5. Serum IGF-1 after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; see also Appendix Table A.13)[1,2,3]



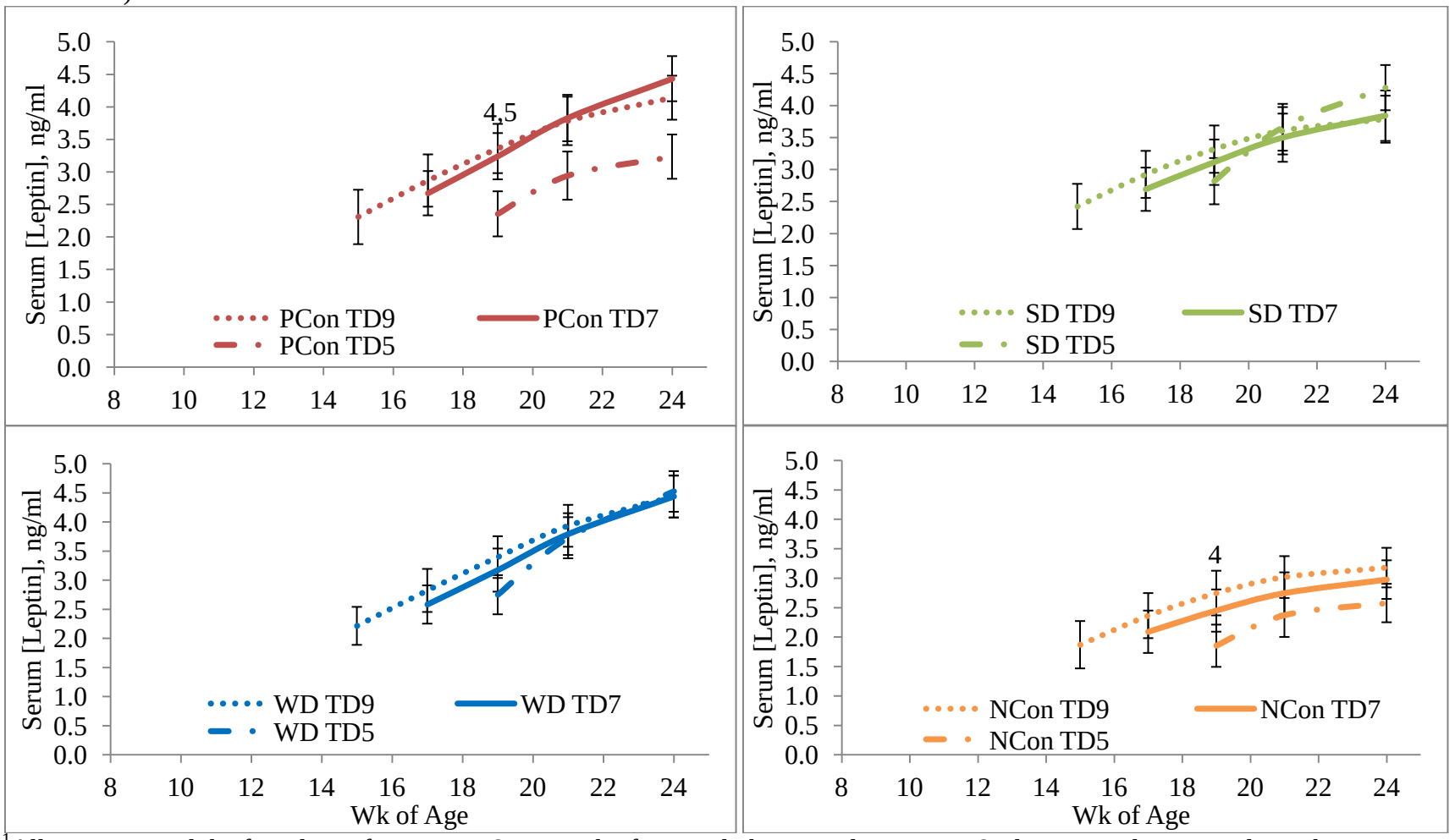
[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P = 0.17$).
[4] Within in each DDGS feeding strategy at each time period, TD5 vs. TD9 ($P < 0.05$).
[5] Within in each DDGS feeding strategy at each time period, TD5 vs. TD7 ($P < 0.05$).

Figure 4.6. Serum IGF-1 after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.13)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P = 0.17$).
[4] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.05$).
[5] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.05$).
[6] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.05$).
[7] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.05$).
[8] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.05$).
[9] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.10$).
[10] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.10$).
[11] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.10$).
[12] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.10$).
[13] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.10$).
[14] Within each Improvest® treatment at each time period, WD vs. SD ($P < 0.10$).

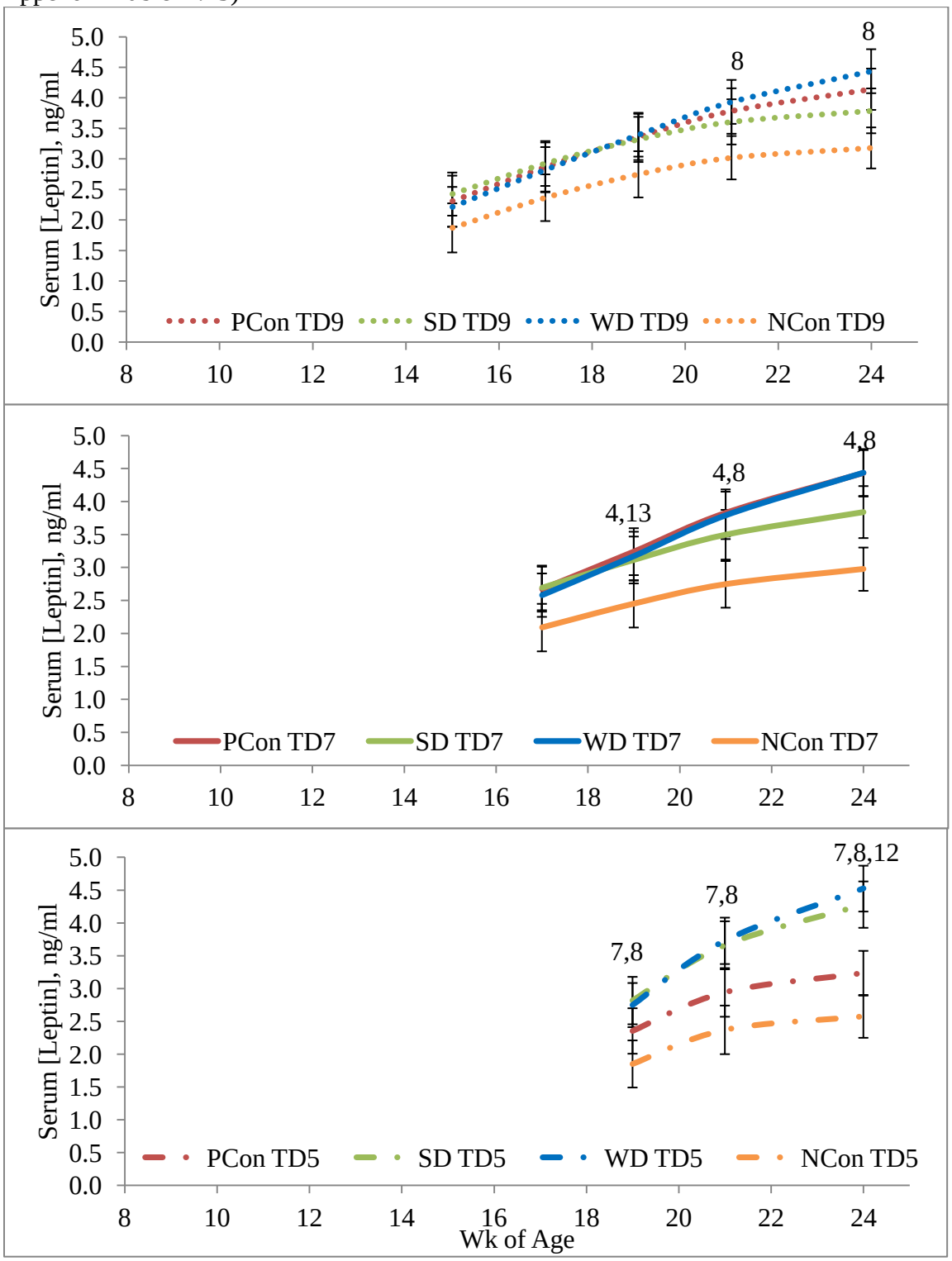
Figure 4.7. Serum leptin after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.13)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or

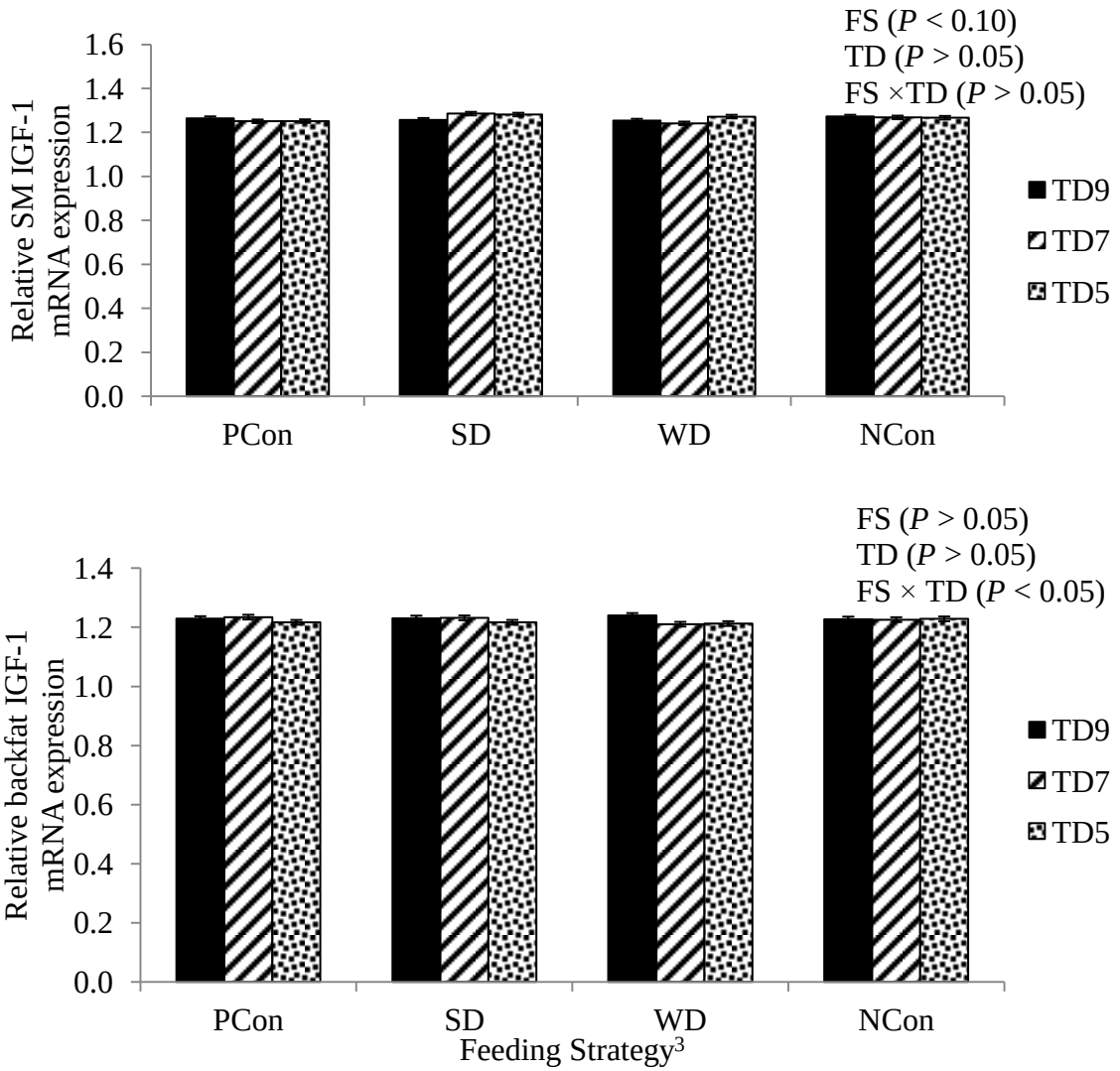
19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P < 0.05$).
[4] Within in each DDGS feeding strategy at each time period, TD5 vs. TD9 ($P < 0.05$).
[5] Within in each DDGS feeding strategy at each time period, TD5 vs. TD7 ($P < 0.05$).

Figure 4.8. Serum leptin after the second dose of Improvest® when immunologically castrated pigs are harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS; See also Appendix Table A.13)[1,2,3]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.
[3] FS × TD × wk^2 ($P < 0.05$).
[4] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.05$).
[5] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.05$).
[6] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.05$).
[7] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.05$).
[8] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.05$).
[9] Within each Improvest® treatment at each time period, SD vs. PCon ($P < 0.10$).
[10] Within each Improvest® treatment at each time period, SD vs. NCon ($P < 0.10$).
[11] Within each Improvest® treatment at each time period, NCon vs. PCon ($P < 0.10$).
[12] Within each Improvest® treatment at each time period, WD vs. PCon ($P < 0.10$).
[13] Within each Improvest® treatment at each time period, WD vs. NCon ($P < 0.10$).

Figure 4.9. Semimembranosus muscle (SM) and backfat adipose mRNA expression of insulin like growth-factor 1 in immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS)[1,2]

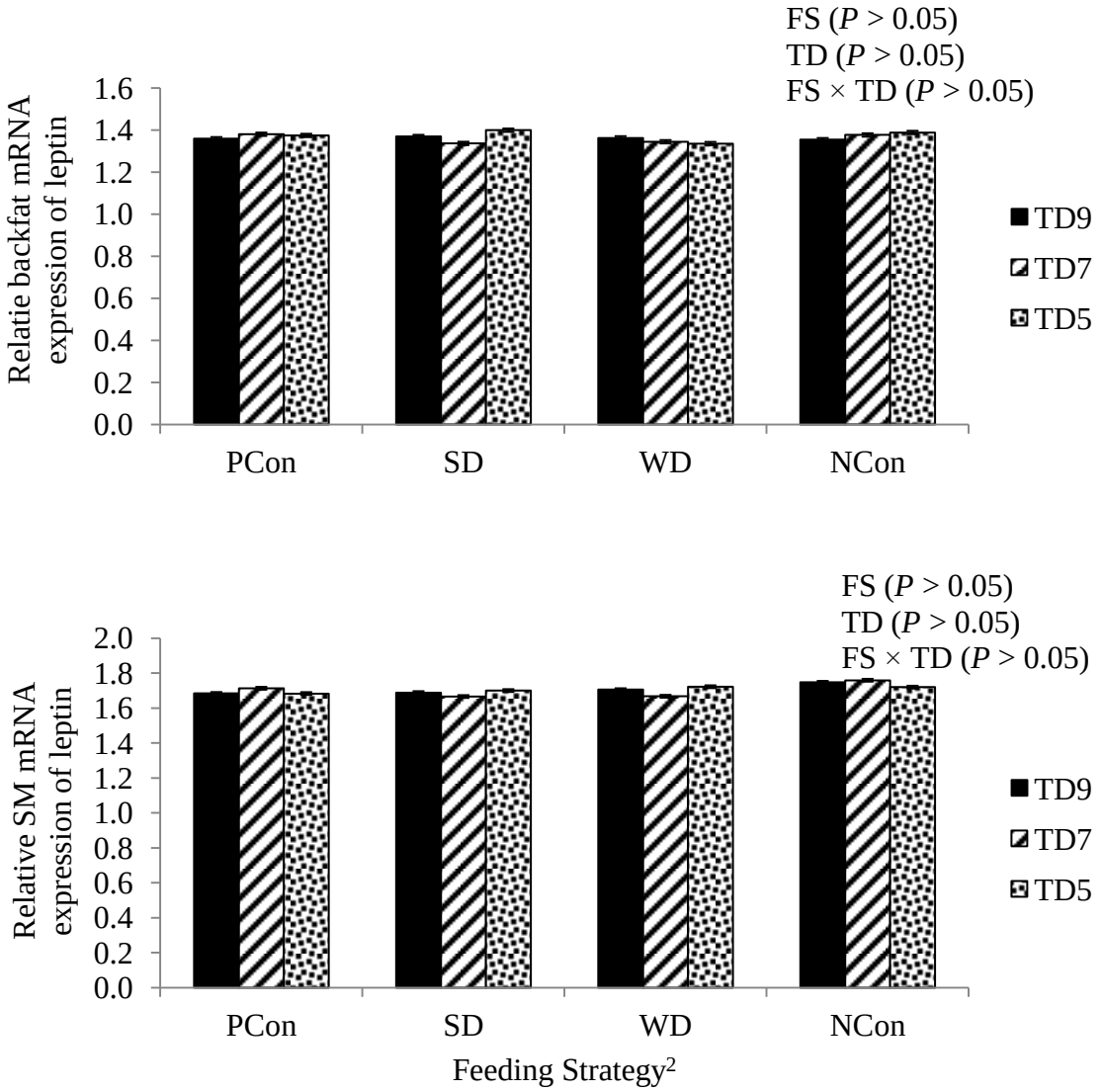


[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] With Tukey adjustment for multiple comparisons there were no significant FS x TD comparisons for backfat IGF-1 mRNA expression or FS of SM IGF-1.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.

Figure 4.10. Semimembranosus muscle (SM) and backfat adipose mRNA expression of leptin in immunologically castrated pigs receiving the second dose at 5, 7, or 9 wk before harvest using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS)[1]



[1]All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] PCon = corn-soybean meal diets containing 0% DDGS throughout growing-finishing period; SD = diets containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; WD = diets containing 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4; NCon = pigs fed diets containing 40% DDGS throughout growing-finishing period.

CHAPTER 5: Effect of time interval between the second Improvest® dose and harvest and DDGS feeding strategies on carcass composition, primal cutout, and pork quality of immunologically castrated pigs

I. Summary

The objective of this study was to evaluate feeding strategies (**FS**) to alleviate the negative effects of feeding diets containing 40% dried distillers grain with solubles (**DDGS**) on carcass composition, primal cutout, and lean quality of immunologically castrated (**IC**) pigs when increasing the interval between the second Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest (**TD**). Intact male pigs (n = 863) were assigned randomly to FS and TD treatments in a 4 × 3 factorial arrangement. Pigs were fed using 1 of 4 FS in a 4-phase feeding program where phases were fed for 3, 4, 4, and 5 wk, respectively. Feeding strategies consisted of: 1) corn-soybean meal (**CS**) control diet with no DDGS (**PCon**), 2) CS + 40% DDGS (NCon), 3) CS + 40, 30, 20, or 10% DDGS in phases 1 to 4, respectively (SD), or 4) CS + 40% DDGS in phase 1 thru 3 and CS in phase 4 (**WD**). Pigs received the second dose of Improvest® at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest. At 24 wk of age (**WOA**), pigs (n = 2/pen) were harvested to determine carcass composition, primal cutout, and lean quality. Pigs fed NCon had reduced ($P < 0.05$) final BW, HCW, chilled carcass wt, and dressing percentage, and had greater ($P < 0.05$) lairage shrink as well as less ($P < 0.05$) ham, whole shoulder, Canadian back loin, and belly primal wt as a percentage of chilled side wt, compared with carcasses

of pigs fed PCon. The WD and SD feeding strategies resulted in loins with intermediate subjective firmness and marbling scores compared with pigs fed PCon and NCon.

Increasing TD resulted in greater ($P < 0.05$) first rib and 10^{th} rib backfat in TD9 and TD7 pigs compared with TD5 pigs. The carcass cutting yield of TD9 pigs tended ($P < 0.10$) to be lower than TD5 pigs, and was mostly attributed to reduced ($P < 0.05$) loin wt of TD9 pigs. Lean quality was not affected by increasing TD.

Pigs fed NCon had reduced carcass dressing percentage, primal yields, and lean quality compared with pigs fed PCon. Use of WD and SD feeding strategies improved carcass dressing percentage and resulted in intermediate primal cut yields and pork loin quality relative to pigs fed PCon and NCon feeding strategies. Increasing the time interval between the second dose of Improvest® and harvest increased adipose tissue accretion but pork lean quality was not different among TD.

KEYWORDS: carcass cutout, dried distillers grains with solubles, feeding strategy, immunological castration, pigs, pork quality

II. Introduction

Pigs immunologically castrated (**IC**) with Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) have reduced carcass fat and improved lean gain efficiency (Squires, 2011), but reduced carcass dressing percentage compared with physically castrated (**PC**) pigs (Boler et al., 2012). These differences between IC and PC pigs have economic implications for pork producers because improved lean gain efficiency reduces dietary cost of gain, but reduced carcass dressing percentage can decrease carcass value due to lower HCW. The time interval between the second Improvest® dose and harvest (**TD**) can range from 3 to

10 wk (FDA, 2011b). A few studies have evaluated the effect of TD at 4 and 6 wk (Boler et al., 2012) and 5 and 7 wk (Tavárez et al., 2014a) on primal cutout and lean quality of IC pigs.

Changes in diet composition (described in Chapter 2) resulting from differences in dietary inclusion rate of dried distillers grains with solubles (**DDGS**), and the rapid increase of ADFI after the second Improvest® dose (Elsbernd et al., 2014), cause changes in nutrient intake which affect BW gain in IC pigs. Feeding diets containing up to 30% DDGS to PC and gilts can result in a linear decrease of dressing percentage (Stein and Shurson, 2009; Leick et al., 2010) and may affect lean quality (Leick et al., 2010; Xu et al., 2010b). Feeding strategies such as removing DDGS before harvest can improve dressing percentage of PC and gilts (Gaines et al., 2007; Xu et al., 2010a), but it is unknown if this feeding strategy (**FS**) would be effective in IC pigs considering the rapid linear increase in ADFI after the second Improvest® dose (Lealiifano et al., 2011). The objectives of this study were to determine the impact of DDGS FS with IC pigs harvested at 5, 7, or 9 wk after the second Improvest® dose on carcass composition, primal cutout, and lean quality, and to identify FS and optimal TD that would mitigate negative impacts on these carcass characteristics.

III. Materials and methods

A. Animals and housing

A detailed description of experimental procedures is found in Chapters 2 and 3. Briefly, intact male (**IM**) pigs (n = 863) were fed for 16 wk at the West Central Research and Outreach Center (Morris, MN) using a 4 × 3 factorial arrangement of treatments which provided 8 replicate pens per treatment combination. Feeding strategies were

evaluated using a 4-phase feeding program for 3, 4, 4, and 5 wk for phase 1 to 4, respectively, and included: positive control (**PCon**) where pigs were fed 0% DDGS corn-soybean meal based diets (**CS**) throughout the growing-finishing period; a DDGS step down (**SD**) feeding strategy where pigs were fed CS containing 40, 30, 20, and 10% DDGS in phases 1 to 4, respectively; a DDGS withdrawal (**WD**) strategy where pigs were fed 40% DDGS in phases 1 to 3, and DDGS was withdrawn from the diet in phase 4 and a CS diet was fed; and a negative control (**NCon**) feeding strategy where pigs were fed diets CS with 40% DDGS throughout the entire growing-finishing period. All pigs had ad libitum access to feed and water. Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) was administered to all pigs at 11 wk of age (**WOA**). The second dose was administered according to treatment assignment at 15, 17, or 19 WOA to correspond with 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) weeks before harvest, respectively.

B. Final weight and dressing percentage

A subsample of pigs (n = 192) were harvested at the University of Minnesota Meat Science Laboratory (St. Paul, MN) for extensive carcass lean and fat quality evaluation. These pigs were selected randomly at 11 WOA. Pigs were weighed and transported 260 km, held in lairage overnight, and provided ad libitum access to water. The following morning, pigs were transported 0.7 km in 2 or 3 groups to the abattoir for harvest. Live BW of pigs was determined before electrical stunning. After stunning, pigs were exsanguinated and scalded to remove hair. Following evisceration, carcasses were split down the midline, and carcasses were weighed with skin and front feet to determine HCW. Following harvest, carcasses were immediately placed in a cooler (mean = 27 min

from stunning). Lairage shrink was determined as the percentage of BW lost between final BW at the barn and final BW before harvest at the abattoir. Dressing percentage was calculated as: final BW before transportation/HCW × 100.

C. Carcass composition and primal cutout

Carcasses were chilled for 24 h at 4°C. The left side of each carcass was separated between the 10^{th} and 11^{th} ribs to determine 10^{th} rib backfat (BF) using a ruler. Acetate tracing paper (TP Orthodontics, Inc; La Porte, IN) was used to trace the LM area. Longissimus muscle area was determined in duplicate by overlaying tracings with a grid. Backfat was also measured along the midline at the first rib, last rib, and last lumbar vertebrae with a ruler. Carcasses were weighed before fabrication to determine chilled carcass weight, and the percentage change from HCW was calculated to determine chilling loss. From the left side of the carcass, heart and leaf fat were removed, and carcasses were fabricated into primals: ham (IMPS #401 and #402), shoulder (IMPS #403, 405, 406), whole belly and IMPS #408, and loin (IMPS #410 and 414). The weight of each primal was calculated as the percentage of the chilled carcass side weight. The IMPS #414 Canadian back loin was used for further pork lean quality assessment. The lean and carcass cutting yields were calculated as described by Boler et al. (2012).

D. Pork quality

The 10^{th} rib location was used to determine pH (Testo model 205, Sparta, NJ) of the longissimus muscle at 45 min and 48 h postmortem. From the anterior end of the Canadian back loin at 48 h postmortem, loins were faced and allowed a minimum of 10 min bloom before objective color was measured with a Hunter colorimeter (MiniScanEZ 4500S; Hunter Lab, Reston, VA; D65 illuminate and 10° observer) at 3 random locations.

Subjective color, marbling, and firmness scores of loins were determined (NPPC, 2000) by a 5-person panel. One loin chop was weighed, suspended from a smoke stick using the fish hook method, and covered with a plastic sealed bag for 24 h (NPPC, 2000). Loin chops were weighed and the percentage of drip loss was calculated. Two loin chops (2.54 cm thick) were vacuum sealed and frozen for later analysis to determine cooking loss and subjective tenderness by instrumental shearing. Frozen chops were thawed, removed from the vacuum package, and initial weight of the chop was recorded. Thermocouplers were inserted to the center of each loin chop, and and each chop was cooked on a clam-style grill to an internal temperature of 70°C. Cooked chops were weighed and percentage of cook loss was calculated. Cooked chops were cooled overnight. Six cores (2.54 cm in diameter) were excised parallel to the muscle fiber orientation and cores were instrumentally sheared (Shimadzu Universal Tester EZ-SX; Kyoto, Japan) perpendicular to the muscle fibers to determine shear force. The remainder of the loin roast was weighed, vacuum sealed, and stored at 4°C for 15 d. Roasts were removed from the vacuum sealed bag, allowed to drip-dry on smoke sticks for 10 min, and weighed to calculate the percentage of roast purge loss.

E. Statistical analysis

The MIXED procedure of SAS (Cary, NC) was used and included fixed effects of DDGS feeding strategy, time of second Improvest® dose before harvest, and the interaction. Pig group was included as a random effect. Hot carcass weight was used as a covariate for carcass measurements of BF and LMA. Least squares means were separated and adjusted using the Tukey option. In instances where normality assumptions of the residuals were not met (subjective marbling and 48 h postmortem pH), data were

transformed using inverse and exponential (-3) transformations, respectively. Means reported for subjective marbling and 48 h postmortem pH have been re-transformed. Differences were considered significant when $P \leq 0.05$ and trends when $P \leq 0.10$.

IV. Results and discussion

A. Effects of DDGS feeding strategy on final BW and carcass dressing percentage

There were no differences in final BW of pigs before transportation among DDGS feeding strategies (Table 5.1). However prior to harvest, the BW of pigs fed NCon was less ($P < 0.05$) than pigs fed PCon (116.3 vs. 121.6 ± 2.7 kg), and pigs fed SD and WD had intermediate ($P > 0.05$) BW compared with pigs fed PCon and NCon. Similarly, pigs fed NCon had less ($P < 0.05$) HCW compared with pigs fed PCon (87.6 vs. 93.1 ± 1.7 kg), and pigs fed NCon tended ($P < 0.10$) to have less HCW compared with pigs fed SD (87.6 vs. 91.4 ± 1.7 kg). These differences in BW before and after harvest were due to a greater ($P < 0.05$) percentage of lairage shrink in pigs fed NCon, and there was a tendency ($P < 0.10$) for pigs fed SD to have a greater percentage of lairage shrink, compared with pigs fed PCon (4.3 and 4.0 vs. 3.4 ± 0.7%, respectively). These differences were also reflected in carcass dressing percentage, where pigs fed NCon had reduced ($P < 0.05$) carcass dressing percentage compared with all other feeding strategies, and pigs fed SD tended ($P < 0.10$) to have reduced carcass dressing percentage compared with pigs fed PCon (73.3 vs. 74.0 ± 0.4%). The differences in BW before harvest, HCW, lairage shrink, and dressing percentage may be attributed to greater gut fill and/or visceral mass due to the relatively higher fiber content of the pigs DDGS diets compared with corn-soybean meal diets (Stein and Shurson, 2009). In addition to greater

gut fill, high fiber diets increase the rate of digesta passage (Kerr and Shurson, 2013) which could have allowed for more rapid digesta excretion when feed was withheld during lairage leading to greater lairage shrink. Both the NCon and SD feeding strategies included diets that contained DDGS up until the final day of feeding (NCon = 40% and SD = 10% DDGS), resulting in higher fiber intake during phase 4 than pigs fed PCon and WD (Chapter 2, Table 6). Use of the SD and WD feeding strategies resulted in similar lairage shrink and dressing percentage (Table 5.1).

B. Effects of DDGS feeding strategy on carcass composition

In the U.S., most packer premium and discount systems place emphasis on HCW. Lean deposition requires less dietary energy than fat deposition (Noblet and van Milgen, 2013). Therefore, it is beneficial to achieve greater HCW by depositing lean rather than adipose tissue. Pigs fed NCon not only had reduced HCW, lairage shrink, and dressing percentage, but tended ($P < 0.10$) to have less carcass first rib backfat compared wih pigs fed the WD feeding strategy (3.33 vs. 3.63 ± 0.10 cm; Table 5.2). The preferential increase in first rib backfat thickness could be due to the increased ME intake of pigs fed the WD feeding strategy after DDGS was removed from the diet in phase 4 (Chapter 3, Figure 2). Excess energy consumption beyond that necessary for lean growth is deposited as fat (Noblet and van Milgen, 2013). The timing of excess energy consumption may have occurred at a time when rate of fat deposition is greatest in the forequarter (first rib location) compared with last rib and last lumbar locations (Apple et al., 2009c). In addition, pigs fed NCon had lower ($P < 0.05$) ME intake during the 15 to 17, 17 to 19, and 21 to 24 wk intervals, compared with pigs fed PCon (Chapter 3, Figure 3.2). Other researchers have observed variable changes in backfat thickness when feeding diets

containing more than 30% DDGS throughout the growing-finishing period. When feeding up to 30% (Xu et al., 2010b) or 45% DDGS (Cromwell et al., 2011), a linear decrease in backfat thickness has been observed. However, others have not observed any difference in backfat thickness when feeding up to 30% (Whitney et al., 2006; Xu et al., 2010a), 40% (Graham et al., 2014), or 60% DDGS (Leick et al., 2010). These discrepancies among studies are difficult to explain because backfat thickness was determined using different instrumentation and at different depot locations. Additionally, feed intake and gain efficiency responses due to dietary inclusion rate of DDGS widely varied among studies. However, the variability in nutrient and energy composition among DDGS sources could have resulted in unexpected changes in nutrient and energy intake in these studies. It is important to note that because dietary fat is deposited more efficiently than carbohydrates (Boisen, 2007), differences in diet nutrient content could have led to the variable changes in backfat thickness.

Changes in energy and nutrient intake can also have consequences on LM area or depth. However, LM measures are more consistent among 5 studies, with only 1 study reporting reduced LM depth when feeding up to 30% DDGS (Whitney et al., 2006). All other studies that have measured LM area or depth have reported no effect of feeding up to 40% (Graham et al., 2014), 45% (Cromwell et al., 2011), or 60% DDGS in diets (Bergstrom et al., 2009a; Leick et al., 2010). Carcasses of pigs fed PCon in the present study tended ($P < 0.10$) to have slightly larger LM area compared with pigs fed SD (43.6 vs. 41.7 ± 0.9 cm^2; Table 5.2). These differences in LM area may have been related to the reduction in overall ME and SID Lys intake of pigs fed SD compared with pigs fed PCon (Chapter 3, Table 3.1). Overall ME intake was also lower in WD and NCon fed pigs

(Chapter 3, Table 3.1) compared with pigs fed PCon, but pigs fed WD and NCon only had numerically smaller carcass LM area compared with pigs fed PCon. Leick et al. (2010) observed a linear decrease in LM depth when feeding up to 60% DDGS in diets to growing-finishing pigs, but growth performance was not determined. These results are in contrast to those reported by Cromwell et al. (2011), who did not observe a difference in LM area when feeding up to 45% DDGS.

Measures of the LM (depth or area), backfat thickness, and HCW, can be combined to calculate carcass fat-free lean percentage. Results from others studies that evaluated effects of feeding DDGS diets to PC and female pigs on fat-free lean percentage are conflicting. Xu et al. (2010b) reported a linear increase in carcass fat-free lean percentage when feeding diets containing up to 30% DDGS, Cromwell et al. (2011) reported a linear trend for greater carcass fat-free lean percentage when feeding diets with up to 45% DDGS, and Bergstrom et al. (2009) reported greater carcass percentage lean when feeding diets containing 60% vs. 20% DDGS. However other studies have observed that feeding up to 30% (Whitney et al., 2006; Xu et al., 2010a), 40% (Graham et al., 2014), or 60% (Leick et al., 2010) DDGS does not affect percentage fat-free lean. This discrepancy could be due to the use of several different equations and instrumentation to measure percentage fat-free lean. Carcass fat-free lean percentage is a general estimate of overall carcass composition but does not reflect the proportion of lean and fat deposition that occurs across primal cuts.

C. Effects of DDGS feeding strategy on primal cut weights

Identifying changes in primal weight is important for evaluating compositional differences among carcasses because the various primal cuts of the pork carcass

contribute different proportions to overall carcass cutout value. Chilled carcass weight was lower ($P < 0.05$) in pigs fed NCon compared with pigs fed PCon (90.2 vs. 84.1 ± 2.4 kg; Table 5.3) due to reduced HCW of pigs fed NCon and the lack of difference in percentage chilling loss. Pigs fed SD and WD had similar chilled carcass weight compared with PCon and NCon fed pigs (86.8 and 86.5 vs. 90.2 and 84.1 ± 2.4 kg, respectively; Table 5.3). The percentage of IMPS 401 and 402 hams relative to chilled carcass wt was reduced ($P < 0.05$) in pigs fed NCon compared with pigs fed PCon. In addition, the IMPS 402 ham weight, as a percentage of chilled side weight, tended ($P < 0.10$) to be less from pigs fed SD and WD compared with pigs fed PCon (24.07 and 24.11 vs. 25.36 ± 0.57%, respectively). The percentage of whole shoulder (IMPS #403) tended ($P < 0.10$) to be lower from pigs fed NCon compared with pigs fed PCon, but when the whole shoulder was split into the picnic (IMPS #405) and butt shoulder (IMPS #406) primal cuts, DDGS feeding strategy did not affect primal cut weights.

Weight of the whole loin as a proportion of chilled carcass side weight was not different, but the trimmed loin (IMPS #410) wt from pigs fed NCon was less ($P < 0.05$), and from pigs fed WD tended ($P < 0.10$) to be less than loin weight from pigs fed PCon (19.94 and 20.24 vs. 21.53 ± 2.55%, respectively). These differences remained when the LM was excised per specification for the IMPS #414. In addition, the Canadian Back loin wt tended ($P < 0.10$) to be reduced from pigs fed SD compared with pigs fed PCon (8.84 vs. 9.49 ± 0.19%). The IMPS specification for the Canadian Back pork loin indicates “all bone, cartilage, tenderloin, and lean and fat over the blade bone shall be removed” (IMPS, 2014) and thus, represents exclusively the LM. Therefore, the differences observed in the Canadian Back loin are representative of changes in lean accretion.

Energy intake is first partitioned toward lean growth and then excess energy is partitioned toward fat deposition (van Milgen and Noblet, 2003). The decreased Canadian Back loin wt from pigs fed NCon, and tendency for decreased loin wt from pigs fed SD and WD compared with pigs fed PCon, can be attributed to differences in ME intake previously described (Chapter 3, Table 3.1). Overall ME intake of pigs fed PCon was greater ($P < 0.05$) compared with pigs fed NCon and SD. (Chapter 3, Table 3.1). Even though pigs fed WD had similar ME intake compared with pigs fed PCon, much of this similarity is due to the rapid increase ($P < 0.05$) of ME intake once the DDGS was removed from the diet in the final 5 wk before harvest, which likely occured after peak lean deposition. Thus, the tendency for reduced Canadian Back loin wt of pigs fed WD is consistent with reduced ME intake of pigs fed WD during phases 1 to 3.

The fresh pork belly (IMPS #408) wt was lower ($P < 0.05$) from pigs fed NCon compared with pigs fed PCon, but bellies from pigs fed the SD and WD strategies had similar weight compared with PCon and NCon pigs (13.07 and 12.87 vs. 13.56 and 12.20 ± 0.95%, respectively; Table 5.3). Collectively, these differences in specific primal cuts did not result in any changes in the lean and carcass yields among DDGS feeding strategies. Other researchers have reported no differences in primal cut weights when growing-finishing pigs were fed diets containing up to 15% DDGS, even though 10th rib backfat and percentage carcass lean decreased linearly with increasing dietary DDGS inclusion rates (Moreno et al., 2010). Detailed primal cutout evaluations from pigs fed DDGS are limited, and most studies reported LM area or depth and backfat thickness as general indicators of pork carcass composition. Carcass primal cutout evaluations have been conducted to determine the effects of dietary energy density. Dietary ME can be

enhanced by adding supplemental fat or reduced by adding fiber (Patience, 2012). Apple et al. (2009) reported that adding 5% supplemental fat from beef tallow, poultry fat, or soybean oil to the diet did not affect primal cut composition even when ME intake was likely different due to greater ME density of fat supplemented diets than the non-supplemented diet (3.35 vs. 3.58 Mcal/kg of diet), and similar ADFI among feeding strategies. Other researchers have reported that increasing dietary ME from 3.30 to 3.48 Mcal/kg of diet reduced the percentage of carcass fat-free lean, but did not alter the percentage lean yield of the ham (Apple et al., 2004). This is in contrast to the findings of the current study where dietary ME content of the phase 4 diets ranged from 3.24 to 3.32 Mcal/kg of diet in PCon and NCon diets, respectively which resulted in reduced in primal ham, loin, and belly primal yields.

D. Effects of DDGS feeding strategy on pork loin quality

Carcass primal composition and quantity of fat and lean are primary influencers of carcass value (Meisinger, 2003). Poor lean quality can result in loss of value due to loss of product weight caused by reduced water-holding capacity and reduced consumer acceptance (Klinkner, 2013). The rate of pH decline is an major determinant of overall lean quality because it affects water-holding capacity, color, and tenderness (Huff-Lonergan et al., 2002). In this study, the LM of pigs fed NCon tended ($P < 0.10$) to have a higher 45 min postmortem pH than pigs fed PCon (6.29 vs. 6.15 ± 0.06; Table 5.5). Greater anaerobic metabolism of glucose leads to greater accumulation of lactic acid in muscle (Aberle et al., 2001), and in theory, reducing the substrate available to produce lactic acid would reduce the pH decline of pork. The CS diets (PCon) fed in this study provided more starch and less fiber and protein than the CS with 40% DDGS diets

(NCon). Low carbohydrate diets have been shown to lower the glycolytic potential of the LM (Rosenvold et al., 2001; Rosenvold et al., 2003), but this did not alter ultimate pH, and may increase shear force (Rosenvold et al., 2001). In this study, the pH of LM was similar among feeding strategies by 48 hr postmortem.

Effects of feeding DDGS diets to PC and gilts on lean color have been variable, with some reporting no effect of feeding up to 30% DDGS (Xu et al., 2010a) or 60% DDGS (Leick et al., 2010) on objective lean color. Researchers who observed changes in lean color when feeding DDGS diets reported a linear decrease in a* and b* values with increasing DDGS (Xu et al., 2010b), but these objective color differences were not great enough to detect using subjective color evaluation as observed in the current study and by Xu et al. (2010b). At 48 h postmortem, the LM had similar L* values, but the LM from pigs fed NCon was less red and less yellow than pigs fed PCon as a result of the lower ($P < 0.05$) a* values (-2.02 vs. -1.51 ± 0.11) and tendency ($P < 0.10$) for lower b* values (5.13 vs. 5.60 ± 0.68, respectively; Table 5.4).

In the present study, the LM from pigs fed SD and NCon had reduced ($P < 0.05$) subjective marbling scores compared with pigs fed PCon (1.23 and 1.21 vs. 1.43 ± 0.04, respectively; Table 5.4). A linear decrease in subjective marbling scores has been observed when feeding diets containing up to 30% (Xu et al., 2010b) or 60% DDGS to pigs (Leick et al., 2010). Intramuscular fat receives the lowest priority for lipid deposition (Aberle et al., 2001). Given that pigs fed NCon had lower ME intake (Chapter 3, Table 3.1), it is likely that there was not enough excess energy consumed to deposit intramuscular fat. By using the WD feeding strategy, subjective marbling was similar to all other feeding strategies, and may have been a result of the dramatic increase in ME

intake after DDGS was removed from the diet in the final five wk of the study (Chapter 3, Figure 3.2).

Subjective firmness of the LM was lower ($P < 0.05$) in pigs fed NCon, and tended ($P < 0.10$) to be lower in pigs fed WD, compared with pigs fed PCon (2.24 and 2.28 vs. 2.60 ± 0.09, respectively; Table 5.4). In general, a reduction in subjective firmness in LM can be associated with reduced water-holding capacity. However, in this study, moisture losses (drip, purge, and cooking) were not affected by DDGS feeding strategy. Results from other studies have shown that feeding diets containing up to 30% DDGS resulted in no change in LM chop drip loss over a 28 d period (Xu et al., 2010b), but the feeding of diets with 30, 45, and 60% DDGS resulted in a greater percentage drip loss compared with pigs fed a standard corn-soybean meal diet (Leick et al., 2010). The reduction in pork fat loin firmness is supported by the inherent increased proportion of phosphoslipids, which have more unsaturated fatty acids compared with triglycerides. (Wood et al., 2008). While not evaluated in this study, pork LM contains less than 2.5% IMF (Klinkner, 2013), and has a higher concentration of unsaturated fatty acids than in belly, jowl, and backfat depots (Wiegand et al., 2011). As a result, an increase in PUFA in the LM of pigs fed NCon and WD may have led to lower subjective firmness scores. A reduction in marbling, lean color, and firmness scores has also been shown to occur when feeding other dietary lipid sources that have less SFA content due to a proportional increase of either MUFA from sunflower oil or PUFA from canola oil (Miller et al., 1990). While chemical composition of the LM was not determined in this study, others have determined fatty acid composition of the LM when feeding diets containing up to 30% DDGS and observed a linear decrease in total MUFA and total SFA, a linear

increase in linoleic and eicosadienoic acids, and greater total PUFA in LM of pigs fed 10, 20, or 30% DDGS compared with pigs fed a corn-soybean meal diet (Xu et al., 2010b). The reduction in pork loin firmness resulting from feeding NCon and WD feeding strategies also suggests that pork fat firmness may also be compromised by these feeding strategies. In fact, pigs fed NCon had greater unsaturated fatty acid content in pork fat and pork bellies were softer (as determined by belly flop distance and flop angle), but feeding the WD strategy improved pork fat firmness compared with pigs fed PCon (Chapter 6). The effect of DDGS feeding strategies on pork fat quality is discussed in greater detail in Chapter 6.

E. Effect of time interval between second Improvest® dose and harvest on final BW weight and carcass dressing percentage

Final BW of pigs before transportation and harvest, and HCW were not different among TD treatments (Table 5.1). However, the percentage lairage shrink was greater ($P < 0.05$) in TD7 and TD5 pigs compared with TD9 pigs (4.0 and 4.3 vs. 3.4 ± 0.7%, respectively) resulting in a tendency ($P < 0.10$) for TD5 pigs to have reduced carcass dressing percentage compared with TD9 pigs (73.0 vs. 73.6 ± 0.4%). Recently, Boler et al. (2014) reported transportation shrink to be greater in IC (harvested 4.5 to 6.5 wk after the second Improvest® dose) pigs compared with PC and IM pigs. Feed intake increases rapidly after the second Improvest® dose (Elsbernd et al., 2014), and likely results in increased gut fill. Boler et al. (2014) reported IC pigs have greater full intestinal tract wt due to greater empty small intestine and stomach wt and numerically greater gut fill compared with PC pigs. The numerical increase in gut fill occurred even after a 12 to 15 h feed withdrawal before harvest. It is possible that differences in gut fill were greater in

IC pigs compared with PC pigs, and that increased fecal excretion during lairage which contributed to greater transportation shrink that was observed by Boler et al. (2014). The greater lairage shrink and reduced dressing percentages (Table 5.1) of TD5 pigs compared with TD9 pigs could be partly due to gut fill in the current study, and appear to be in agreement with observations by Boler et al. (2014). Additionally, pigs that are harvested with shorter time intervals between the second Improvest® dose and harvest have larger testes (Lealiifano et al., 2011). This would also explain the reduced dressing percentage of TD5 pigs compared with TD9 pigs in the present study. However, changes in reproductive tract mass do not explain the changes in lairage shrink and it is unlikely that larger testes, and presumably reproductive tracts, accounted for all of the reduction in dressing percentage among TD treatments.

F. Effects of time interval between second Improvest® dose and harvest on carcass composition

Greater ($P < 0.05$) carcass BF depth was observed for TD9 pigs at the 10th rib (1.83 vs. 1.67 ± 0.05 cm), and tended ($P < 0.10$) to be greater at the first rib (3.57 vs. 3.34 ± 0.10 cm), and last lumbar BF (1.69 vs. 1.52 ± 0.08 cm) than TD5 pigs. At the last lumbar, TD9 pigs fed NCon tended ($P < 0.10$) to have more BF than TD5 pigs fed NCon (1.86 vs. 1.42 ± 0.13 cm; Figure 5.1). There was no difference in BF thickness between TD9 and TD7 pigs at any location. In most studies, IC pigs have less BF than PC pigs (Dunshea et al., 2001; Pauly et al., 2009; Boler et al., 2012; Yuan et al., 2012), or tend to have less BF than PC pigs (Asmus et al., 2014b). Lealiifano et al. (2011) reported a linear increase in BF thickness when increasing the interval between the second Improvest® dose and harvest from 0 to 6 wk. However, Boler et al. (2012) reported no change in BF

thickness when increasing the interval from 4 to 6 wk, and Asmus et al. (2014) reported no change in BF thickness when increasing the interval from 5 to 7 wk. These differences among studies may be due to differences in HCW where pigs harvested 6 wk after the second Improvest® dose weighed 71 kg (Lealiifano et al., 2011), and HCW of pigs in the studies by Boler et al. (2012) and Asmus et al. (2014) were greater than 90 kg. The reduced BF thickness in IC pigs compared with PC pigs at heavier HCW was also observed by Boler et al. (2014), where HCW exceeded 100 kg. This would suggest that backfat deposition not only increases after the second Improvest® dose, but increases more rapidly when the time interval between the second Improvest® dose and harvest is increased. This has practical implications in the U.S. pork industry where HCW of pigs exceeds 95 kg. To capture the typical reduction in BF thickness of IC pigs compared with PC pigs, the second Improvest® dose would need to be delayed when marketing pigs at HCW exceeding 100 kg.

Increasing the time interval between the second Improvest® dose and harvest did not affect carcass LM area. This result is consistent with results from other studies where carcass LM area was similar between IC and PC pigs (Batorek et al., 2012; Yuan et al., 2012; Asmus et al., 2014b; Boler et al., 2014; Batorek et al., 2012; Yuan et al., 2012). Depth of the LM has been reported to be less (Boler et al., 2014), or tends to be less (Boler et al., 2012) in IC pigs compared with PC pigs when using the Fat-O-Meter, but was not different between IC and PC pigs when using other methods to determine LM depth (Batorek et al., 2012; Asmus et al., 2014b). Furthermore, increasing the time interval between the second Improvest® dose and harvest has been shown to have no effect on LM area or depth (Boler et al., 2012, Asmus et al., 2014b).

G. Effects of time interval between second Improvest® dose and harvest on primal cut weights

The only difference in carcass primal weights among Improvest® treatment times was for loins, where TD9 pigs tended ($P < 0.10$) to have a greater percentage of whole loin weight compared with TD7 pigs (31.74 vs. 30.17 ± 0.91%), but TD5 pigs had similar whole loin weight compared with TD9 and TD7 pigs (Table 5.3). Standardizing the whole loins to IMPS 410 involves trimming loins to 0.10 cm of subcutaneous fat, but this did not change the loin weight difference between TD9 and TD5 pigs. However, TD5 pigs had greater ($P < 0.05$) loin weight, and TD9 pigs tended ($P < 0.10$) to have heavier loins, compared with TD7 pigs (21.07 and 20.75 vs. 19.76 ± 2.54%, respectively). Weights of Canadian back loins were not different among TD treatments. This finding is in contrast to results reported by Boler et al. (2012), where whole and trimmed loin weight did not differ between immunologically castrated pigs harvested at 4 and 6 wk after the second Improvest® dose, but Canadian back loins were heavier in IC pigs harvested 4 wk after the second Improvest® dose compared with IC pigs harvested 6 wk after the second Improvest® dose.

The differences in loin weights, and lack of differences in all other primals resulted in a tendency ($P < 0.10$) for TD5 pigs to have greater carcass cutting yield percentage than TD9 pigs (69.6 vs. 68.7 ± 2.65%). While IC increases lean and carcass cutting yields, increasing the time between second Improvest® dose and harvest from 4 to 6 wk did not alter lean or carcass cutting yields (Boler et al., 2012)

H. Effect of time interval between second Improvest® dose and harvest on pork loin quality

Increasing the time from 5 to 9 wk between the second Improvest® dose and harvest did not affect any measure of pork loin quality in this study. Only 1 other study evaluated pork loin quality of IC pigs harvested a 4 and 6 wk after the second Improvest® dose. Boler et al. (2012) observed greater LM a* and b* values, lower subjective marbling and firmness scores, and greater percentage drip loss in IC pigs harvested 6 wk after the second Improvest® dose compared with IC pigs harvested 4 wk after the second Improvest® dose (Boler et al., 2012). The reduced LM firmness scores of IC pigs harvested 6 wk after the second Improvest® dose may have been related to lower subjective marbling scores and greater percentage drip loss (Boler et al., 2012). The observed increase in LM a* by Boler et al. (2012), and the lack of differences among Improvest® treatments in the current study, could also be function of weight and age. Boler et al (2012) extended the time interval between the second Improvest® and harvest by increasing pig age, whereas in the current study, age of the pig was held constant. Latorre et al. (2004) reported a linear increase in a* value (more red) and myoglobin with increasing BW at harvest.

In this study, the interaction between DDGS feeding strategy and the time interval between the second Improvest® dose and harvest had minimal effects on body composition, primal cutout, and pork lean quality. Feeding diets containing 40% DDGS to IC pigs reduced lean deposition due to decreased HCW, carcass LM area, and primal cutout. Increasing the time interval between second Improvest® dose and harvest increased carcass fat in TD9 pigs compared with carcass fat of TD5 pigs. Lean quality of

pork was unaffected by the timing of the second Improvest® dose relative to harvest, but feeding 40% DDGS diets resulted in softer pork loins with less marbling. While there is no industry standard for loin firmness or marbling, softer loins may need to undergo additional processing steps, such as freezing, to make them amenable for slicing loin chops. Use of the WD feeding strategy improved marbling, but not LM firmness, whereas the SD feeding strategy improved firmness, but not marbling, compared with pigs fed the PCon feeding strategy. The diet related reductions in LM firmness and marbling could be indicative of quality traits connected with reduced pork fat quality, which is of concern due to the high linoleic acid content of corn oil in DDGS. This is of greater concern due to the lower backfat thickness of TD5 pigs compared with TD9 pigs. Pigs with less backfat typically are more sensitive to dietary fatty acid composition. Therefore, the interaction between DDGS feeding strategy and time interval between the second Improvest® dose and harvest on the effect of pork fat quality needs to be evaluated.

Table 5.1. Body weight before transportation and prior to harvest, hot carcass weight, lairage shrink, and carcass dressing percentages of immunologically castrated pigs harvested at 5, 7, or 9 weeks before harvest and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Table A.14)

	Feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value[3]	
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD
Final BW before transport, kg	125.9	124.3	122.2	121.4	2.7	123.3	123.2	123.8	2.6	0.11	0.94
Final BW at harvest, kg	121.6^{a}	119.3^{ab}	117.4^{ab}	116.3^{b}	2.6	119.0	118.5	118.5	2.5	0.04	0.93
HCW, kg	93.1^{a}	91.4^{ax}	89.7^{ab}	87.6^{by}	1.7	90.7	90.3	90.3	1.6	< 0.01	0.93
Lairage shrink[4], %	3.4^{ax}	4.0^{by}	3.7^{ab}	4.3^{b}	0.7	3.4^{a}	4.0^{b}	4.3^{b}	0.7	< 0.01	< 0.01
Carcass dressing[5], %	74.0^{ax}	73.3^{by}	73.5^{ab}	72.2^{c}	0.4	73.6^{x}	73.1	73.0^{y}	0.4	< 0.01	0.08

[1]PCon = pigs fed 0% DDGS, corn-soybean meal diets throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and a 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at 15, 17, or 19 wk of age corresponding to 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively, before harvest.

[3] FS × TD ($P \geq 0.72$).

[4]Lairage shrink loss percentage calculated as [(final BW before transport - BW before harvest)/ final BW before transport] × 100.

[5]Dressing percentage calculated as (final BW before transport/ HCW) × 100.

[a,b,c] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.10$).

Table 5.2. Carcass backfat thickness and LM area of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Table A.15)

	Feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD
Backfat thickness[3], cm												
First rib	3.60	3.51	3.63^{x}	3.33^{y}	0.10	3.57^{ax}	3.64^{a}	3.34^{by}	0.10	0.07	0.02	0.82
Last rib	2.17	2.14	2.23	2.14	0.10	2.23	2.16	2.12	0.10	0.70	0.34	0.33
Last lumbar	1.60	1.61	1.59	1.61	0.08	1.69^{x}	1.60	1.52^{y}	0.08	1.00	0.07	0.08
10th rib	1.75	1.81	1.80	1.73	0.05	1.83^{a}	1.82^{ax}	1.67^{by}	0.05	0.71	0.03	0.46
LM area[3], cm^2	43.6^{x}	41.5^{y}	41.7	42.4	0.9	42.3	42.5	42.2	0.9	0.07	0.94	0.82

[1]PCon = pigs fed 0% DDGS, corn-soybean meal diets throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and a 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at 15, 17, or 19 wk of age corresponding to 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively, before harvest.

[3] HCW was used as a covariate ($P \leq 0.01$).

[a,b] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.10$).

Table 5.3. Chilling loss, carcass primal weights, and lean and carcass cutting yields of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Tables A.16 and A.17*

	DDGS feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value[3]	
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD
Chilled carcass wt, kg	90.2^{a}	86.8^{ab}	86.5^{ab}	84.1^{b}	2.4	88.3	85.2	87.2	2.3	< 0.01	0.12
Chilling loss, %	3.2	3.1	3.1	3.2	0.3	3.1	3.2	3.1	0.3	0.80	0.76
Internal fat (heart and leaf fat), % chilled side wt	2.32	2.32	2.45	2.41	0.11	2.49	2.39	2.24	0.10	0.72	0.11
IMPS, % chilled side wt											
401 Ham	28.40^{ax}	27.11^{ab}	26.91^{by}	26.43^{b}	0.64	27.32	26.76	27.56	0.61	0.01	0.28
402 Trimmed ham	25.36^{ax}	24.07^{by}	24.11^{by}	24.64^{b}	0.57	24.38	23.76	24.64	0.54	< 0.01	0.14
403 Whole shoulder	23.81^{x}	23.48	23.19	22.41^{y}	0.70	23.59	22.62	23.46	0.66	0.10	0.11
405 Picnic shoulder	11.20	10.42	10.68	10.33	0.68	10.76	10.41	11.03	0.67	0.13	0.14
406 Butt shoulder	11.17	11.12	10.79	10.43	1.15	11.04	10.74	10.86	1.14	0.17	0.65
Whole loin	31.81	30.99	30.69	30.31	0.95	31.74^{x}	30.17^{y}	30.94	0.91	0.31	0.09
410 Loin	21.53^{ax}	20.40^{ab}	20.24^{by}	19.94^{b}	2.55	20.75^{ax}	19.76^{by}	21.07^{a}	2.54	0.02	0.02
414 Canadian Back loin	9.49^{ax}	8.84^{by}	8.85^{by}	8.68^{b}	0.19	9.01	8.72	9.16	0.17	0.02	0.15
408 Belly	13.56^{a}	13.07^{ab}	12.87^{ab}	12.20^{b}	0.95	13.10	12.71	12.97	0.93	0.04	0.62
Lean cutting yield[4], %	57.74	57.99	57.59	57.96	3.02	57.53	57.54	58.39	3.01	0.85	0.12
Carcass cutting yield[5], %	69.13	69.30	68.79	68.87	2.66	68.71^{x}	68.76	69.60^{y}	2.65	0.71	0.07

*n = 6 observations (pens) per treatment combination.

[1]PCon = pigs fed 0% DDGS, standard corn-soybean meal diet throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS in 4 dietary phases, respectively; WD = pigs fed 40% DDGS in phases 1 - 3 and 0% DDGS in phase 4; NCon = pigs fed 40% DDGS throughout growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age; Second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.

[3] FS × TD; ($P > 0.10$).

[4] [(picnic shoulder + butt shoulder + loin + ham primal cut wt)/ chilled carcass wt] × 100; (Boler et al., 2012).

[5] [(picnic shoulder + butt shoulder + loin + ham + belly primal cut wt)/ chilled carcass wt] × 100; (Boler et al., 2012).

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

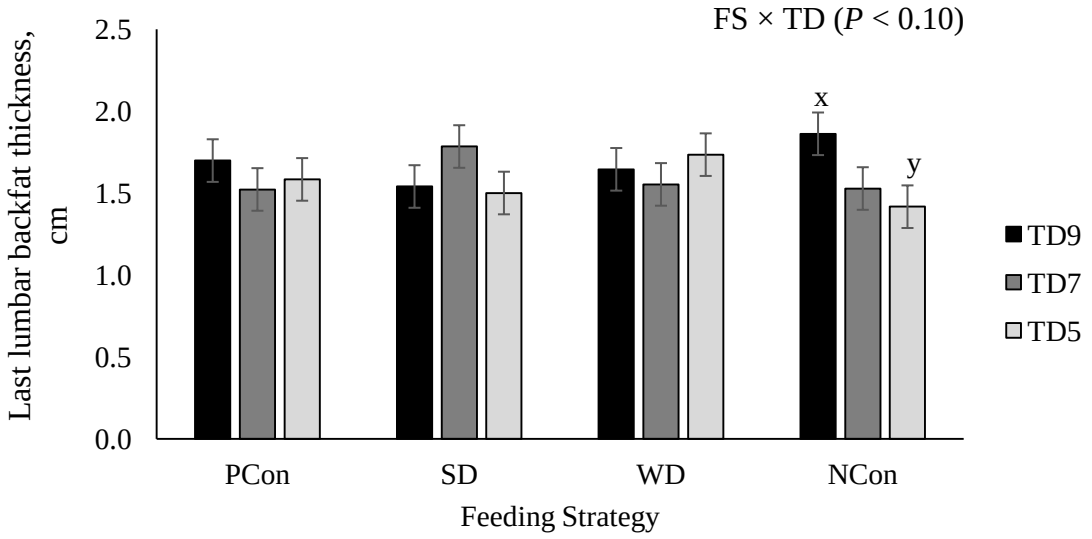
[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Table 5.4. Objective and subjective pork loin quality assessment of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and using 4 different corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Table A.18)

	DDGS feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD
pH 45 min	6.15[x]	6.20	6.18	6.29[y]	0.06	6.20	6.20	6.21	0.06	0.08	0.93	0.22
pH 48 h[3]	5.65	5.70	5.66	5.69	0.05	5.70	5.66	5.67	0.04	0.48	0.48	0.41
LM L*	45.8	45.2	44.2	44.4	2.5	44.7	44.9	45.1	2.5	0.11	0.79	0.70
LM a*	-1.51[a]	-1.76[ab]	-1.77[ab]	-2.02[b]	0.11	-1.65	-1.76	-1.88	0.10	0.02	0.21	0.20
LM b*	5.60[x]	5.39	5.19	5.13[y]	0.68	5.39	5.35	5.24	0.67	0.09	0.65	0.43
Subjective color[4]	2.44	2.50	2.41	2.27	0.09	2.48	2.42	2.30	0.08	0.29	0.23	0.42
Subjective marbling[4]	1.43[a]	1.23[b]	1.30[ab]	1.21[b]	0.04	1.28	1.29	1.28	0.04	< 0.01	0.99	0.95
Subjective firmness[3,4]	2.60[ax]	2.32[ab]	2.28[by]	2.24[b]	0.09	2.44	2.28	2.36	0.08	0.01	0.32	0.82
Roast purge loss[3], %	1.87	1.44	1.88	1.94	0.25	1.88	1.81	1.66	0.24	0.10	0.50	0.37
Drip loss, %	2.18	1.78	2.02	1.77	0.29	1.88	2.10	1.83	0.29	0.14	0.28	0.69
Cook loss, %	18.9	19.5	18.5	18.2	1.0	18.6	18.9	18.8	0.9	0.35	0.83	0.05
Shear force, kg	3.09	3.15	3.22	3.03	0.14	3.11	3.17	3.08	0.12	0.80	0.88	0.89

[1]PCon = pigs fed 0% DDGS, standard corn-soybean meal diet throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS in 4 dietary phases, respectively; WD = pigs fed 40% DDGS in phases 1 - 3 and 0% DDGS in phase 4; NCon = pigs fed 40% DDGS throughout growing-finishing period.
[2] All pigs received the first dose of Improvest® at 11 wk of age; Second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.
[3] For subjective marbling and 48 h postmortem pH, data were analyzed and transformed using an inverse and exponential (-3) transformations, respectively. Reported means have been re-transformed.
[4] (NPPC, 2000)
[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 5.1. Interactive effect of immunologically castrated pigs receiving the second dose of Improvest® at 5, 7, or 9 weeks before harvest and using 4 corn dried distillers grains with solubles (DDGS) feeding strategies (FS) on carcass backfat thickness at the last lumbar location[1, 2, 3]



[1]PCon = pigs fed 0% DDGS, corn-soybean meal diets throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and a 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.
[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at 15, 17, or 19 wk of age corresponding to 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively, before harvest.
[3] HCW was used as a covariate ($P \leq 0.001$).
[x,y] Means without a common superscript differ ($P \leq 0.10$).

CHAPTER 6: Effectiveness of using different corn dried distillers grains with solubles feeding strategies and increasing the time intervals between the second Improvest® dose and harvest of immunologically castrated pigs on belly and pork fat quality

I. Summary

To determine the effectiveness of corn dried distillers grains with solubles (**DDGS**) feeding strategies (**FS**) when increasing time interval between the second Improvest® dose and harvest (**TD**), pigs were fed corn-soybean (**CS**) meal control diet (**PCon**), CS + 40, 30, 20, or 10% DDGS in 4 phases, respectively (**SD**), CS + 40% DDGS in phases 1 to 3 and CS 5 wk before harvest (**WD**), or CS + 40% DDGS diets (**NCon**) throughout the growing-finishing period. Pigs received the second Improvest® dose at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest. Overall, linoleic acid intake was similar between pigs fed SD and WD (45.4 vs. 50.0 ± 2.6 g/d), which was intermediate ($P < 0.05$) compared with pigs fed NCon and PCon (64.4 and 27.7 ± 2.6 g/d, respectively). Overall, linoleic acid intake of TD9 and TD7 pigs was similar, but greater ($P < 0.05$) than TD5 pigs (47.7 and 47.7 vs. 46.0 ± 2.4 g/d, respectively). Bellies tended ($P < 0.10$) to be thinner and belly fat iodine value (IV) was greater ($P < 0.05$) in TD5 compared with TD9 pigs (67.2 vs. 65.8 ± 1.2, respectively). Linoleic acid content was similar in all carcass fat depots of pigs fed NCon (> 22.7%). Pigs fed PCon had greater ($P < 0.05$) linoleic acid content in jowl fat compared with belly and backfat depots (13.2 vs. 11.4 and 10.6 ± 0.9%, respectively). Jowl fat of pigs fed WD had greater ($P < 0.05$) linoleic acid content than all other fat depots from pigs fed SD, WD, and PCon. Use of the Meadus et al. (2010) IV equation consistently resulted in higher IV than the AOCS (1998) IV equation

thus the difference between these equations (IV-diff) was calculated. Jowl fat of pigs fed NCon (3.59 ± 0.11) had greater ($P < 0.05$) IV-diff than all other FS and fat depots. Backfat and belly fat of pigs fed NCon had similar IV-diff compared with jowl fat of pigs fed SD and WD (3.08, 3.14, 3.05, and 3.16 ± 0.11, respectively). In conclusion, increasing TD reduced IV in all fat depots and increased pork belly thickness. However in all TD treatments, IV of all fat depots were lower than commonly used IV thresholds for acceptable pork fat quality. The WD and SD feeding strategies were effective in reducing IV in all fat depots. Use of the Meadus et al. (2010) IV equation is more sensitive for assessing pork fat quality than the AOCS (1998) because it includes more long-chain unsaturated fatty acids that may lead to softer pork fat and greater susceptibility to peroxidation.

KEYWORDS: belly quality, dried distillers grains with solubles, feeding strategy, immunological castration, pigs, pork fat

II. Introduction

Intact male (**IM**) pigs have less backfat with higher concentrations of PUFAs than gilts (Wood et al., 1989). Pork fat that contains high concentrations of PUFAs is soft and undesirable (Hugo and Roodt, 2007). Immunologically castrated (**IC**) pigs can be harvested between 3 to 10 wk after the second Improvest® dose (FDA, 2011a). Increasing the interval between the second Improvest® (*gonadotropin releasing factor analog - diphtheria toxoid conjugate*; Zoetis, Inc, Florham Park, NJ) dose and harvest (**TD**) up to 6 wk, results in a linear increase in backfat thickness (Lealiifano et al., 2011). Thus, IC pigs harvested with shorter TD are predisposed to have fat depots with greater unsaturated fatty acid content. In belly fat, high concentrations of PUFA are of particular

concern because of poor handling and processing characteristics of soft pork bellies (Morgan et al., 1994). Belly fat iodine value (**IV**) can be reduced by increasing the TD (Asmus et al., 2014b; Boler et al., 2014). Dietary lipid sources rich in PUFA, such as corn dried distillers grains with solubles (**DDGS**; NRC, 2012), increase IV of jowl, belly, and backfat (Leick et al., 2010; Cromwell et al., 2011) as dietary inclusion rate increases. When withdrawing 30% DDGS from the diet, IC pigs have a greater decrease in belly fat IV than physical castrates (**PC**) (Asmus et al., 2014b). However, at times it is economically favorable, to include more than 30% DDGS to growing-finishing pig diets, and other DDGS feeding strategies (**FS**) may be used. The effect of various DDGS FS on belly and pork fat quality in IC pigs has not been evaluated. Additionally, multiple IV equations exist that may alter interpretation of acceptable pork fat quality. Therefore, the objectives of this study were to determine the effectiveness of using DDGS withdrawal (**WD**) and stepdown (**SD**) FS in IC pigs harvested at TD5, TD7, or TD9 to improve belly and pork fat quality, and to compare the use of 2 common IV equations for assessing pork fat quality.

III. Materials and methods

A. Animals and housing

Carcass fat samples used in this study were collected from IC pigs that were fed various DDGS feeding strategies previously described in Chapter 2. Briefly, intact male pigs (n = 863) were randomly assigned at 8 wk of age (**WOA**) to 1 of 4 DDGS feeding strategies and time between the second Improvest® dose and harvest in a 4 × 3 factorial arrangement of treatments. Experimental diets were fed in 4 dietary phases representing 3, 4, 4, and 5 wk, respectively. Feeding strategies included feeding 0% DDGS throughout

the entire growing-finishing period (**PCon**), decreasing dietary DDGS inclusion from 40, 30, 20, and 10% DDGS in phase 1 to 4, respectively (**SD**); feeding 40% DDGS in phases 1 to 3 and 0% DDGS in phase 4 (**WD**); or feeding 40% DDGS throughout the growing-finishing period (**NCon**). All pigs were administered the first dose of Improvest® at 11 WOA, and the second dose was administered at 15, 17, or 19 WOA. All pigs were harvested at 24 WOA which corresponds to pigs receiving the second dose of Improvest® at 9 (**TD9**), 7 (**TD7**), or 5 (**TD5**) wk before harvest. These selected time intervals between the second Improvest® dose and harvest fit within the approved 3 to 10 wk time period when IC pigs can be harvested (FDA, 2011a).

B. Dietary lipid and linoleic acid intake

Feed disappearance was determined at the beginning and end of each dietary phase, and 2 wk after the beginning of phases 2 to 4 (Chapter 2). Feed offered during each feeding period was recorded and the feed remaining at the end of each feeding period was subtracted from the total feed offered to calculate feed disappearance. For each feeding period, lipid intake was calculated as (total feed intake × analyzed diet lipid content)/(pigs per pen × d on feed). Linoleic acid intake was calculated for each feeding period by [(total feed intake × (analyzed diet lipid content × analyzed linoleic acid content of lipid)]/(pigs per pen × d on feed).

C. Harvest and sample collection

At 13 WOA, a subsample of pigs (n = 2 pigs/pen) were selected randomly to be harvested at the University of Minnesota Meat Science Laboratory (St. Paul, MN) to assess belly and pork fat quality. Harvest and carcass fabrication occurred as described in Chapter 5. During fabrication, subcutaneous carcass fat from the jowl and 10th rib

backfat along the midline were collected. The belly (IMPS #408) was chilled for 24 h after fabrication at 4°C.

D. Quality assessment and fatty acid analysis

The length and width of each belly was measured by ruler at the mid-point. Belly thickness was determined at 4 locations along the dorsal and 4 locations along the ventral edges. Bellies were draped skin-side down over a smoke stick and the distance between skin-to-skin anterior and posterior ends was measured with a ruler. The belly flop angle was calculated as $\cos^{-1}[0.5(L^2) - D^2]/[0.5(L^2)]$, where L = belly length and D = skin-side down, skin-to-skin belly distance (Whitney et al., 2006). Carcass fat from the anterior dorsal corner of the belly was collected after morphometric assessment. Objective Hunter (MiniScanEZ 4500S; Hunter Lab, Reston, VA; D65 illuminate and 10° observer) color values of carcass fat from each depot were determined immediately after sample collection. Subjective Japanese Color Score (JCS; 1 = white and 4 = yellow) of belly samples was evaluated by a single observer. All carcass fat samples were immediately frozen at -20°C for further analysis following morphometric and color assessment.

Fatty acid analysis of carcass fat samples was conducted by the University of Missouri Agricultural Experiment Station Chemical Laboratories (Columbia, MO). Total SFA was calculated as the summation of C8:0, C10:0, C12:0, C14:0, C15:0, C16:0, C18:0, C20:0, C21:0, C22:0, C23:0, and C24:0. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in carcass fat samples. Total MUFA were calculated as the summation of C16:1n-6 palmitoleic, C17:1n-10, C18:1n-9t elaidic, C18:1n-9 oleic, C18:1n-11 vaccenic, C20:1n-9 gonodic, C22:1n-9 erucic, C24:1n-9 nervonic acids, and total PUFA was calculated as the summation of C18:2t linoelaidic, C18:2n-6 linoleic,

C18:3n-3 linolenic, C20:2, C20:2n-3 homo-γ-linolenic, C20:4n-6 arachidonic, C22:2n-6, C20:5n-3 eicosapentaenoic (EPA), C22:4n-6 adrenic, C22:5n-3clupanodonic, and C22:6n-3 docosahexaenoic (DHA) acids. Total n3 fatty acids included C18:3n-3, C20:2n-3, C20:5n-3, C22:5n-3, and C22:6n-3, and total n6 fatty acids included C18:2n-6, C20:3n-6, C20:4n-6, C22:2n-6, C22:4n-6. Iodine value was calculated using AOCS, 1998) and Meadus et al., 2010) equations (Table 6.1), and the difference in IV between these equations (IV-diff) was also calculated.

E. Statistical analysis

Data were analyzed using Proc MIXED in SAS (Cary, NC) where FS and TD treatments, carcass fat depot, and all interactions were included as fixed effects. The replication group was included as a random effect. Hot carcass weight was used as a covariate ($P < 0.01$) for belly length, width, thickness, and flop distance. The residuals were used to test the assumptions of normality using PROC Univariate. When the Kolmogorov-Smirnov test for normality was significant ($P < 0.05$), any datapoint with a residual > ± 4, was removed from analysis. Least squares means were separated and the Tukey adjustment was used.

IV. Results and discussion

Typically, pork fat is predominantly composed of oleic acid (Wood et al., 2008), but fatty acid composition can vary greatly, depending on dietary fatty acid composition, sex of the pig (Wood et al., 1989), BW (Apple et al., 2009a), and carcass fat depot (Apple et al., 2011; Duttlinger et al., 2012). Fatty acids deposited in adipose tissue originate from two sources, either *de novo* lipogenesis, where fatty acids are synthesized from carbohydrate metabolism, or fatty acids are derived directly from dietary lipids (White et

al., 2013). Enzymatic activity that catalyzes the synthesis of fatty acids through *de novo* lipogenesis decreases at varying rates during the growing-finishing period, with jowl fat having the lowest lipogenic activity and belly fat having the greatest lipogenic activity in pigs at 120 kg BW (Mourot et al., 1995). Thus, the predominance of fatty acid deposition derived from the diet is greater in jowl fat compared with belly fat, and occurs later in the finishing phase.

The effects of dietary lipid sources and dietary fatty acid composition have been studied for decades (Ellis and Isbell, 1926). It is widely accepted that feeding highly unsaturated fatty acid sources results in pork fat that is also relatively unsaturated (Miller et al., 1990; Apple et al., 2009b). The melting temperature of fatty acids decreases as fatty acids become more unsaturated which results in soft pork fat (Wood et al., 2004). Poor pork fat quality has been described as soft, oily, wet, grey, and floppy (Wood et al., 1992). Intact male pigs have less backfat than gilts, and the fatty acid composition of pigs with less backfat is more unsaturated (Wood et al., 1989). Since pigs with less backfat have carcass fat that is more unsaturated, these pigs are more sensitive to dietary changes in fatty acid composition. In gilts and PC, feeding diets containing DDGS results in an increase in unsaturated fatty acids in pork fat compared with feeding corn-soybean meal diets. For example, feeding diets containing 30% (Xu et al., 2010b, Cromwell et al., 2011) and 45% (Cromwell et al., 2011) DDGS generally doubles the linoleic acid content of belly fat (Xu et al., 2010b) and backfat (Cromwell et al., 2011) compared with pigs fed standard corn soybean meal diets. One solution to overcome soft pork fat is to minimize dietary linoleic acid intake.

A. Lipid and fatty acid intake

Like corn, the predominant fatty acid in DDGS is linoleic acid, but DDGS has a greater lipid content than corn (3.5% vs. 5 to 14%, respectively; NRC, 2012). The source of DDGS used in this study contained 10.4% ether extract and the ether extract contained 53.9% linoleic acid (Chapter 2, Tables 2.4 and 2.5). In addition to the lower lipogenic activity that occurs late in the finishing phase, linoleic acid suppresses Δ9 desaturase which minimizes the abundance of oleic acid, and increases the proportion of linoleic acid in carcass fat depots (Kouba and Mourot, 1998).

The current study evaluated 2 commonly used DDGS feeding strategies (WD and SD) in IC pigs, which have been used for PC and gilts. There was no interaction between FS × TD × wk for lipid or linoleic acid intake (Figure 6.1 and 6.2, Appendix, Table A.20). In phase 1 of this study, SD, WD, and NCon fed pigs had similar lipid and linoleic acid intake since they were all fed 40% DDGS diets. However, the gradual decline of DDGS throughout phases 2 thru 4 for pigs fed SD resulted in greater ($P < 0.05$) linoleic acid intake compared with pigs fed PCon throughout phases 2 to 4, but lower ($P < 0.05$) linoleic acid intake compared with pigs fed NCon and WD during phases 2 and 3. During phase 4, pigs fed SD (10% DDGS) had greater ($P < 0.05$) linoleic acid intake compared with WD and PCon (0% DDGS). As expected, pigs fed the WD feeding strategy had a dramatic decrease ($P < 0.05$) in lipid and linoleic acid intake during the 19 to 21 wk feeding interval compared with the 17 to 19 wk feeding interval (lipid intake was 72.9 vs. 132.9 ± 5.55 g/d, respectively; linoleic acid intake was 39.30 vs. 71.95 ± 2.83 g/d, respectively) once DDGS was removed from the diet at 19 WOA (Figure 6.1 and 6.2). These changes in linoleic acid intake throughout the growing-finishing period

resulted in similar overall linoleic acid intake between pigs fed the SD and WD feeding strategies, and markedly reduced ($P < 0.05$) overall linoleic acid intake compared with pigs fed NCon (45.4 and 50.0 vs. 65.4 ± 2.6 g/d; Table 6.1). In the current study, pigs fed PCon had an average daily linoleic acid intake of 27.7 g/d, which was much lower than the 46.7 g/d linoleic acid intake of pigs fed a corn-soybean meal diet reported by Kellner (2014). This can be attributed to lower ADFI and lower dietary lipid concentration in all 4 dietary phases in the current study compared with diets fed by Kellner (2014). Differences in ADFI between the present study and those reported by Kellner (2014) are not surprising since Kellner (2014) used PC and gilts and the current study used IC pigs, which typically have lower ADFI compared with PC and gilts (Puls et al., 2014).

Changes in lipid and linoleic acid intake among TD treatments were the result of changes in ADFI (Figure 2.3). During the 17 to 19 wk feeding interval, TD9 pigs had greater ($P < 0.05$) lipid and linoleic acid intake than TD7 and TD5 pigs (linoleic acid: 61.4 vs. 55.3 and 51.6 ± 2.56 g/d, respectively; Figure 6.2 and Appendix Table A.21). During the 19 to 21 wk feeding interval, TD9 and TD7 pigs had greater ($P < 0.05$) linoleic acid intake than TD5 pigs (56.3 and 59.1 vs. 49.5 ± 2.6 g/d, respectively). The rapid increase in ADFI of TD5 pigs during the final 3 wk feeding period (Figure 2.3) resulted in a rapid increase in linoleic acid intake so that linoleic acid intake was greater ($P < 0.05$) in TD5 than TD9 pigs (61.6 vs. 55.8 ± 2.6 g/d, respectively). Recent work describing the relationship between fatty acid intake and carcass fatty acid composition, showed that minimizing linoleic acid intake before harvest is essential for minimizing carcass fat depot IV (Kellner, 2014). Immunologically castrated pigs have less backfat than PC (Dunshea et al., 2001; Boler et al., 2012), and as a result, are a greater concern

for having carcass fat that contains high concentrations of unsaturated fatty acids. Furthermore, IC pigs also have reduced ADFI, which leads to pigs in the current study having less linoleic acid intake compared with PC and gilts reported by Kellner (2014), when fed a corn-soybean meal diet. On the other hand, controlling linoleic acid intake of TD5 pigs by using diets or FS that reduce unsaturated fatty acid intake is of greater importance after the second Improvest® dose because of the rapid increase in feed and linoleic acid intake immediately before harvest, as well as less backfat of TD5 pigs compared with TD7 and TD9 pigs (Chapter 5, Table 5.2).

B. Belly characteristics

Belly quality has received increased attention due to increased consumer demand and value of bacon, coupled with the relatively high fat content and greater fat:lean (D'Souza et al., 2004) compared with other pork carcass primals. Soft pork bellies can lead to challenges in handling, may cause reductions in slicing yield, have an unattractive appearance, and are more susceptible to oxidation and off-flavor development (Wood et al., 2008). Thicker bellies increase slicing yield and quality, but decrease visual acceptability and intent to purchase for consumers (Person et al., 2005). As a result, morphometric belly parameters including thickness, flop distance, and flop angle are commonly measured to assess belly quality. Additionally, some consumers use adipose tissue color as an indicator of pork quality, and prefer bright white fat (Hugo and Roodt, 2007). Determination of fatty acid composition requires collecting belly samples, which causes deformation and devaluation of high-value belly primals. Therefore, other carcass fat depots have been sought as alternative predictors of belly quality.

In the current study, there were no FS × TD interaction of morphometric belly measures (Table 6.2 and Appendix Table A.22). Pigs fed NCon had lower ($P < 0.05$) HCW and reduced ($P < 0.05$) percentage of belly primal weight in the chilled carcass compared with pigs fed PCon (HCW: 85.3 vs. 90.6 ± 1.5 kg and belly primal weight percentage: 12.20 vs. 13.56 ± 0.95%). Pigs fed SD and WD tended ($P < 0.10$) to have lower HCW compared with pigs fed PCon (88.3 and 87.8 vs. 90.6 ± 1.5 kg, respectively), but the percentage of chilled carcass belly primal weight from pigs fed the SD and WD feeding strategies was not different compared with pigs fed PCon and NCon (13.07 and 12.87 vs. 13.56 and 12.20 ± 0.95%, respectively). Compared with pigs fed PCon, bellies from pigs fed NCon were thinner ($P < 0.05$; 2.97 vs. 2.80 ± 0.20 cm, respectively) with reduced firmness as indicated by the reduced ($P < 0.05$) flop distance (8.66 vs. 5.57 ± 0.65 cm, respectively) and flop angle (16.4 vs. 10.3 ± 1.4°, respectively). The decrease in belly weight, as a percentage of chilled carcass weight, and reduced belly thickness of pigs fed NCon may have been due to the reduced ME intake (Chapter 3, Table 3.1) and ADG (Chapter 2, Table 2.8) of these pigs compared with those fed other FS. The reduced flop distance and flop angle of pigs fed NCon in this study is similar to values reported by others, where belly angle, flex score, and flop distance have linearly decreased due to feeding diets containing up to 30% (Whitney et al., 2006, Xu et al., 2010b), 45% (McClelland et al., 2012), or 60% DDGS (Leick et al., 2010), and demonstrate that feeding increasing levels of DDGS results in softer pork bellies in gilts and PC.

Both SD and WD feeding strategies restored belly thickness similar ($P > 0.05$) to pigs fed PCon (2.95 and 2.98 vs. 2.97 ± 0.20 cm, respectively). Flop distance and angle were improved ($P < 0.05$) in pigs fed SD and WD compared with pigs fed NCon

(distance: 7.34 and 7.11 vs. 5.57 ± cm; angle: 14.0 and 13.4 vs. 10.3 ± 1.4°), but flop distance of pigs fed SD and WD was lower ($P < 0.05$) compared with pigs fed PCon. Flop angle was lower ($P < 0.05$) in pigs fed WD, and tended ($P < 0.10$) to be lower in pigs fed SD compared with pigs fed PCon (14.0 and 13.4 vs. 16.4 ± 1.4°, respectively). However, Xu et al., 2010a) reported no changes in morphometric softness or belly thickness by withdrawing 30% DDGS from the diet for up to 9 wk, which may have been a result of bellies being unusually thin (< 1.63 cm) compared with those evaluated in other studies, where belly thickness was > 2.71 cm, > 3.18 cm, and > 3.0 cm when feeding up to 30% (Whitney et al., 2006), 45% (McClelland et al., 2012), and 60% (Leick et al., 2010) DDGS diets, respectively. Responses to dietary-induced changes in belly thickness have been variable, where a linear decrease has been observed when feeding up to 30% (Whitney et al., 2006) or 60% DDGS (Leick et al., 2010) diets, resulting in a 14 to 17% decline in belly thickness. However, other researchers have reported no change in belly thickness when feeding up to 30% (Xu et al., 2010b) or 45% DDGS diets (McClelland et al., 2012).

One objective of increasing the time interval between the second Improvest® dose and harvest was to increase belly thickness, since backfat thickness increases linearly with increasing the time interval post-second dose of Improvest® and harvest (Lealiifano et al., 2011). This is important because increasing belly thickness increases bacon slicing yield (Person et al., 2005). In this study, increasing the time interval between the second Improvest® dose and harvest tended ($P < 0.10$) to increase belly thickness of TD9 pigs compared with TD5 pigs (2.99 vs. 2.86 ± 0.93 cm, respectively). Interestingly, Boler et al. (2012) reported thinner bellies in IC pigs harvested 6 wk after

the second Improvest® dose compared with pigs harvested 4 wk after the second dose. Tavárez et al., 2014a) observed that harvesting IC pigs at 5 wk after the second Improvest® dose resulted in thinner bellies with less flop distance compared with PC pigs of the same age. Immunologically castrated pigs harvested 7 wk after the second dose, tended to have thinner bellies, but flop distance was similar compared with PC pigs of the same age. Thus, the effect of IC on belly thickness and flop distance was greater with shorter intervals between the second Improvest® and harvest (Tavárez et al., 2014a). However, withdrawing 30% DDGS from the diet 7 wk before harvest improved belly thickness to levels similar to pigs fed PCon when harvested 7 wk after the second Improvest® dose (Tavárez et al., 2014a). There was no effect of feeding 30% DDGS diets or withdrawing 30% DDGS from the diet in pigs harvested 5 wk after the second Improvest® dose on belly thickness or flop distance (Tavárez et al., 2014a). This observation is in contrast to the theory that pigs with shorter TD are more sensitive to dietary changes in dietary fatty acid intake. However, it is possible that the pigs harvested 7 wk after the second Improvest dose benefited more from the withdrawal feeding strategy because the withdrawal period was 2 wk longer than pigs harvested 5 wk after the second Improvest dose. From the description of study design of Tavárez et al., 2014a), it is difficult to separate the effects related to time interval between the second dose and harvest, and the length of the DDGS withdrawal period.

Regardless of FS or TD treatment, the belly flop distance, angle, and thickness reported in this study were lower than those reported by Whitney et al. (2006) and Xu et al. (2010) in PC and gilts, and those reported in PC and IC pigs (Boler et al., 2012, Kyle et al., 2014, Tavárez et al., 2014a). However, bellies obtained in this study would have

been classified as "average" thickness (2.5 cm) as described by Person et al. (2005). Person et al. (2005) determined that the processing yield and slicing yield of #1 slices was greater in "thick" (3.0 cm) bellies compared with "average" (2.5 cm) and "thin" (2.0 cm) bellies. However, for consumers, bacon slices from "thick" bellies was less visually acceptable compared with bacon from bellies of "average" (2.5 cm) or "thin" (2.0 cm) thickness. Therefore, Person et al. (2005) recommended that pork producers focus on producing pigs with bellies averaging 2.5 cm thick as a compromise to accommodate consumer visual preference for bacon from thinner bellies, and greater processing and slicing yields of thicker bellies.

C. Carcass fat color

In some international markets, such as the Japanese market, consumers prefer a bright white fat color (Morgan et al., 1995). Dried distillers grains with solubles is derived from corn, which is an abundant source of carotenoids. However, about 25 to 50% of the carotenoid activity is lost due to drying of DDGS (Winkler-Moser, 2012). Variation in drying time and temperature used by ethanol plants to produce DDGS, along with variable dietary inclusion rates for DDGS, could lead to variable changes in pork fat color. In a study where diets containing up to 30% DDGS were fed to PC and gilts, backfat, but not belly fat L* and b* linearly decreased (Xu et al., 2010b), and when diets containing up to 60% DDGS were fed, belly fat L* decreased linearly (Leick et al., 2010). These objective color changes in carcass fat depots resulted in subjective color score differences in belly or backfat depots (Xu et al., 2010b).

In the current study, objective jowl and backfat color were unaffected by DDGS feeding strategy (Table 6.3). However, feeding NCon resulted in darker ($P < 0.05$) belly

fat color compared with feeding PCon and SD, as indicated in lower L* value (76.5 vs. 78.6 and 78.1 ± 0.6, respectively). Withdrawing DDGS 5 wk before harvest resulted in intermediate ($P > 0.05$) belly fat L* compared with PCon, SD, and NCon (77.6 vs. 78.6, 78.1, and 76.5 ± 0.6, respectively) feeding strategies. Belly a* and b* were unaffected by DDGS feeding strategy.

While the changes in L* observed in this were study were small, these differences were also observed in the subjective evaluation of belly adipose color using the JCS scale. Pigs fed NCon had higher ($P < 0.05$) JCS compared with pigs fed PCon and SD, (1.33 vs. 1.08 and 1.15 ± 0.07, respectively) and pigs fed WD tended ($P < 0.10$) to have greater JCS compared with pigs fed PCon (1.29 vs. 1.08 ± 0.07, respectively). While changes in belly adipose color were detected by both objective and subjective evaluation, it is unknown if these differences would be considered unacceptable for consumers. The variable differences in objective color among studies and depots could be due to the variation in carotenoid content among DDGS sources resulting from different drying methods. It could be that carotenoids are preferential deposited with specific fatty acids given that fatty acid composition also differs among carcass fat depots. Another possibility is that depots that have less lipid filling would have a greater proportion of connective tissues which have more pigmentation.

D. Fatty acid composition and iodine value of pork carcass fat depots

Backfat of IM pigs has a more unsaturated fatty acid profile than gilts (Wood et al., 1989), and backfat (Benz et al., 2010) as well as belly fat (Correa et al., 2008) from gilts has a more unsaturated fatty acid profile than PC. Pork fat becomes more saturated with increasing backfat thickness (Wood et al., 1989). In fact, in an unidentified

subcutaneous fat depot, IC pigs had intermediate linoleic acid, total SFA and PUFA content compared with PC and IM pigs (Pauly et al., 2009). While IC pigs have similar expression of fatty acid synthase and steroyl co-a desaturase compared with PC, IC pigs have greater Δ6 desaturase activity compared with PC which results in belly fat with a more unsaturated fatty acid content due to fatty acid desaturation and elongation (Mackay et al., 2013). Immunologically castrated pigs can be harvested between 3 to 10 wk after the second dose of Improvest® (FDA, 2011a), which results in an increase in feed intake (Lealiifano et al., 2011, Elsbernd et al., 2014) and backfat thickness as the time interval increases (Lealiifano et al., 2011). Therefore, increasing the time interval between the second dose of Improvest® and harvest could lead to greater saturated fatty acid content in carcass fat.

E. Effect of increasing the time interval between the second Improvest® dose and harvest on fatty acid composition and iodine value of pork carcass fat depots

Previous research results have shown that increasing the time interval between the second dose of Improvest® and harvest from 4 to 6 wk decreased linoleic acid content and tended to decrease iodine value of belly fat (Boler et al., 2012). Similarly, linoleic acid content of belly fat declined when increasing the interval between the second Improvest® dose and harvest from 2 to 8 wk (Tavárez et al., 2014b). Others have reported that increasing the interval between the second dose and harvest resulted in a greater decrease of carcass fat IV in IC pigs compared with PC pigs, but jowl fat was less responsive to increasing the time interval than belly fat and backfat (Asmus et al., 2014b). Tavarez et al. (2014) observed a greater jowl fat IV in IC pigs harvested 2 wk

after the second Improvest® dose compared with PC pigs of the same age. When IC pigs were harvested 4 wk after the second Improvest® dose, jowl fat IV of IC pigs tended to be greater than PC pigs, but IC pigs harvested at 6 and 8 wk after the second Improvest® dose had similar jowl fat IV to PC pigs of the same age. However, in these studies, the greater intervals between the second dose of Improvest® and harvest were achieved by increasing the pig age, and the increased saturated fatty acid content of carcass fat of IC pigs also occurred in PC of the same age.

Other researchers have achieved an increase in backfat thickness when maintaining the same interval between the second Improvest® dose and harvest, but increasing age and BW of pigs (Dunshea et al., 2001). However, under these conditions, changes in backfat fatty acid composition are unknown. The increased saturated fatty acid content of belly fat from IC and PC pigs observed by Boler et al. (2012) and Tavárez et al., 2014b) may have been more related to increasing backfat thickness by increasing the age and BW (physiological age) rather than by increasing TD (chronological age). Increasing BW typically increases saturated fatty acid content of adipose tissue in PC and gilts as shown by the decreasing iodine value of backfat from 64 to 80.5 kg HCW (Apple et al., 2009a). Other researchers have harvested pigs with 80, 86, and 94 kg HCW and observed no change in fatty acid composition of belly fat (Correa et al., 2008). In both cases, as backfat thickness increased, oleic acid content increased and linoleic acid decreased (Wood et al., 1989). As a result, an increase in saturated fatty acid content in carcass fat may be limited at heavier BW. The increase in saturated fatty acids in carcass fat with increasing BW did not occur when feeding a diet high in PUFA provided by the addition of 5% soybean oil to the diet (Apple et al., 2009a).

To understand the impact of increasing the interval between the second Improvest® dose and harvest, animal age must be kept constant as was done in the current study. Reducing the time interval between the second Improvest® dose and harvest increased ($P < 0.05$) the linoleic and linolenic acid content in jowl, backfat, and belly fat of TD5 pigs compared with TD7 and TD9 pigs (linoleic: 17.8 vs. 16.8 and 16.7 ± 0.8%, respectively; linolenic: 0.75 vs. 0.70 and 0.71 ± 0.04%, respectively; Table 6.5). In addition, reducing the time interval between the second Improvest® dose and harvest tended ($P < 0.10$) to increase the arachidonic acid content of TD5 pigs compared with TD9 pigs (0.41 vs. 0.39 ± 0.01%, respectively) in all carcass fat depots. The increase in linoleic and linolenic acid content in all adipose depots of TD5 compared with TD7 pigs occurred when palmitic acid tended ($P < 0.10$) to be reduced in TD5 pigs compared with TD7 pigs (22.9 vs. 23.3 ± 0.2%; Table 6.5). Subsequently, across all 3 carcass fat depots, total SFA tended ($P < 0.10$) to be greater in TD9 than TD5 pigs (36.8 vs. 36.0 ± 0.4%), while total PUFA was greater ($P < 0.05$) in TD5 than TD7 and TD9 pigs (19.9 vs. 18.8 and 18.8 ± 0.9%). These main effect differences in fatty acid composition of all 3 fat depots are likely due to altering the timing of the second Improvest® dose and harvest which directly affected changes in ADFI and linoleic acid intake.

F. Effect of DDGS feeding strategy on fatty acid content of carcass fat depots

In the current study, linoleic acid content was not different among jowl, belly, and backfat from pigs fed the NCon feeding strategy (Table 6.4). However, pigs fed the PCon strategy had greater ($P < 0.05$) linoleic acid content in jowl fat compared with the backfat and belly fat (13.2 vs. 10.6 and 11.4 ± 0.9%). For pigs fed the NCon feeding strategy, the concentration of linolenic, eicosadienoic, and arachidonic acids were greater ($P < 0.05$) in

jowl fat compared with backfat and belly fat. The increase in unsaturated fatty acid content in carcass fat depots when feeding NCon (40% DDGS) to IC pigs is similar to the responses reported by other researchers when feeding 45% DDGS diets to PC and gilts (McClelland et al., 2012). Increasing the diet inclusion rate of DDGS up to 60% resulted in a linear increase in linoleic acid content in the belly and jowl depots at the expense of a linear decrease in oleic, stearic, and palmitic acids in the jowl, and a linear decrease in oleic acid in belly fat (Leick et al., 2010). Similarly, McClelland et al. (2012) observed a linear increase in linoleic and eicosadienoic acid content of backfat and belly fat, a linear increase in linolenic acid in the belly fat, and a linear increase in arachidonic acid in backfat at the expense of a linear decrease in oleic, stearic, and palmitic acids when increasing dietary DDGS inclusion.

The SD and WD feeding strategies were effective at reducing ($P < 0.05$) the linoleic acid content in all 3 carcass fat depots relative to pigs fed NCon. However, the SD and WD feeding strategies resulted in greater ($P < 0.05$) linoleic acid content in all carcass fat depots compared with pigs fed PCon. Pigs fed the WD feeding strategy had greater ($P < 0.05$) linoleic acid content in jowl fat than in belly fat, and belly fat had greater ($P < 0.05$) linoleic acid content than backfat (19.4 vs. 17.1 vs. 14.7 ± 0.9%, respectively). However, pigs fed PCon and SD feeding strategies had similar linoleic acid content in backfat and belly fat, and both depots had less ($P < 0.05$) linoleic acid content than jowl fat. Relative to the NCon feeding strategy, the SD and WD feeding strategies were also effective at reducing ($P < 0.05$) the content of linolenic and eicosadienoic acids in the jowl and backfat depots, but only the SD feeding strategy reduced ($P < 0.05$) the content of linolenic and eicosadienoic acids in the belly fat. These reductions resulted in

linolenic and eicosadienoic acid concentrations that were similar to pigs fed PCon, regardless of feeding strategy and fat depot. Arachidonic acid was unchanged in backfat and belly fat regardless of feeding strategy. In jowl fat, the SD and WD feeding strategies had no effect on arachidonic acid content, but feeding the PCon strategy resulted in lower ($P < 0.05$) arachidonic acid compared with pigs fed NCon.

The variable increases in PUFA content among carcass fat depots came at the expense of reducing ($P < 0.05$) palmitic, stearic, and oleic acids in all carcass fat depots. The SD and WD feeding strategies resulted in similar total SFA, MUFA, and PUFA content within a given depot. Total PUFA content was greater in the jowl than in the belly and backfat depots for all FS. Interestingly, FS had a similar effect on total PUFA content in belly fat and backfat, except that pigs fed the WD feeding strategy had greater ($P < 0.05$) total PUFA in the belly fat than in backfat. In fact, TD7 and TD5 pigs were less responsive to the WD feeding strategy for lowering dietary linolenic acid content, and linolenic acid content was similar to pigs fed the NCon strategy (Figure 6.3). Linolenic acid content of belly fat from TD9 pigs fed the WD strategy was lower ($P < 0.05$) compared with the NCon feeding strategy, but similar compared with pigs fed the SD and PCon strategies (0.652 vs. 0.774, 0.647, and 0.624 ± 0.053%, respectively; Figure 6.3). Total n3 fatty acids were greater in all carcass fat depots from pigs fed NCon compared with all other feeding strategies, and were greater in jowl fat of pigs fed NCon compared with backfat and belly fat (1.12 vs. 0.96 and 0.90 ± 0.05%; Table 6.4).

G. Comparison of iodine value of carcass fat depots using different equations

Iodine value has been used as a composite value representing the proportion of unsaturated fatty acids in adipose tissue, where a higher value represents greater

unsaturated fatty acid content and softer, less desirable pork fat. The most commonly reported IV is calculated using an equation established by the AOCS (1998), and places greatest emphasis, by using larger coefficients, on linolenic acid followed by linoleic, palmitoleic, oleic, gondoic, and erucic acids (Table 6.6; AOCS, 1998). It is important to note that 2 different IV equations have been used to evaluate the unsaturated fatty acid content of pork fat depots (AOCS, 1998; Meadus et al., 2010). The AOCS (1998) IV equation does not include fatty acids greater than 20 and 22 carbons long, except for gondoic and erucic acids, while the Meadus et al. (2010) equation includes these fatty acids (Table 6.6). Using both the AOCS and Meadus equations resulted in similar relative IV differences among DDGS feeding strategies and TD treatments in the current study, but using the Meadus equation consistently resulted in higher IV, ranging from 2.68 to 3.59 units. Most of the increase in the Meadus IV was due to including eicosadienoic and arachidonic acids in the calculation.

The incremental IV increase of the Meadus equation varied among feeding strategies and carcass fat depots. The IV-diff of pigs fed NCon was greater ($P < 0.05$) than all other DDGS feeding strategies within a given carcass fat depot, especially in jowl fat (Table 6.4). Jowl fat from pigs fed the NCon and WD strategies had greater ($P < 0.05$) IV-diff than backfat and belly fat of pigs provided the same FS. Pigs fed PCon and SD strategies had similar IV-diff among fat depots. Small quantities of linolenic, eicosatrienoic, and arachidonic acids are present in pork carcass fat depots. Even though these fatty acids are in relatively low concentrations, the chemical properties of these fatty acids negatively affect pork fat quality. Increasing the number of interrupted methylenes (e.g. double bonds) in fatty acids lowers the melting point and exponentially

reduces the dissociative energy necessary for hydrogen abstraction to occur (Erickson, 2008). Thus, fatty acids with more double bonds are more easily peroxidized.

Additionally, fat quality plays a role in the overall quality of lean meat. Arachidonic and linoleic acids are the major fatty acids in muscle (Wood et al., 2008). Arachidonic acid contains 6 double bonds and thus is easily peroxidized. However, arachidonic acid is not included in the IV equation established by the AOCS (1998), but is included in the Meadus et al. (2010) equation, thus, arachidonic acid accounts for a major portion of IV-diff. Additionally, most of the long chain fatty acids accounted for in IV-diff were not present in the diet (Chapter 2, Tables 2.6 and 2.7). As a result, they must have been synthesized from linoleic and linolenic acids. If the objective of using IV is to identify compositional differences that result in bellies with poor handling and processing characteristics, as well as, bellies that are more susceptible to peroxidation and product deterioration, then greater consideration should be given to more long-chain fatty acids. Use of the Meadus IV equation may be more advantageous for classifying belly and pork fat quality as determined by fatty acid composition and IV.

H. Improving and predicting pork fat quality

Several strategies have been developed to improve and predict pork fat quality. In addition to using SD and WD strategies, formulating diets based on iodine value product (IVP) is effective for managing dietary effects on carcass IV until saturated fatty acids are included in "high" IVP diets (Benz et al., 2011a). This led Benz et al. (2011) to suggest that IVP is a poor predictor of carcass fat IV when using different fatty acid sources to formulate diets to a similar IV. Benz et al. (2011) suggested that linoleic acid intake was a better predictor of carcass fat IV. Other researchers have also observed that the addition

of animal fats with high saturated fatty acid content to diets containing high concentrations of unsaturated fatty acids results is no effect, or minimal improvements in saturated fatty acid content of pork fat depots (Pomerenke et al., 2011; Lee et al., 2013;). The use of IV as a measure of pork fat quality has been questioned because other aspects of fatty acid profiles may be more useful (McClelland et al., 2012). Equations using linoleic acid intake (Averette Gatlin et al., 2002; Kellner, 2014), or dietary DDGS inclusion (Cromwell et al., 2011) as predictors of jowl, belly, and backfat IV have been published, and more recently, equations have been developed to account for intake of essential fatty acids and length of feeding (Paulk et al., 2014). Length of feeding maybe an important factor to consider in predicting carcass fat fatty acid composition because deterioration of pork fat quality occurs very quickly (Apple et al., 2009a; Browne et al., 2013) when feeding dietary fats with high levels of unsaturated fatty acids. However, improving pork fat quality is more difficult by removing dietary ingredients containing high concentrations of unsaturated fatty acids. Strategies such as removing or changing the fat sources or adding saturated dietary fat sources (Pomerenke et al., 2011; Browne et al., 2013; Lee et al., 2013) appear to only be effective when increasing the length of time for feeding these types of diets (Xu et al., 2010a; Browne et al., 2013)

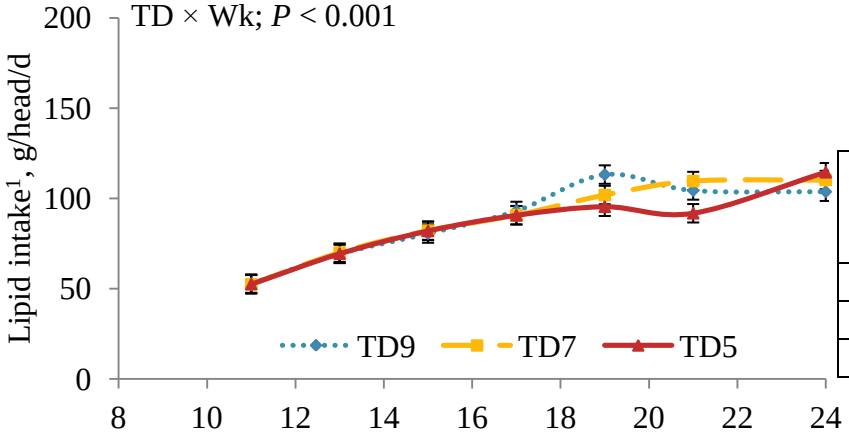
In conclusion, increasing the time interval between the second Improvest® dose and harvest resulted in slight changes in belly quality and carcass fatty acid composition, with a tendency for increased belly thickness for pigs harvested at 9 wk compared with 5 wk after the second Improvest® dose, and no changes in adipose tissue color or belly flop. Increasing the time interval from 5 to 7 wk decreased IV of all carcass fat depots, but IV was unchanged when increasing the time interval from 7 to 9 wk after the second

Improvest® dose. However, for all TD treatments, IV was below commonly accepted IV thresholds that range from 70 (Barton-Gade, 1987) to 75 (Boyd et al., 1997) when using either the AOCS or Meadus equations. Feeding strategy had a much greater impact on carcass fatty acid composition and belly quality than TD treatments. As expected, pigs fed 40% DDGS diets throughout the growing-finishing period had greater carcass fat linoleic acid content and IV compared with all other feeding strategies. The use of SD and WD feeding strategies were effective in reducing linoleic acid content and IV in all 3 fat depots. However, linoleic acid content and IV of jowl, belly, and backfat from pigs fed SD and WD were greater than pigs fed PCon. Relative to belly and backfat from pigs fed NCon, linolenic acid content was reduced in backfat, but not belly fat, in TD5 and TD7 pigs fed WD. However, in TD9 pigs, linoleic acid content of pigs fed WD was reduced in backfat and belly fat compared with TD9 pigs fed NCon. This inconsistent response in fatty acid composition of jowl, belly, and backfat depots to DDGS feeding strategies underscores the inability to use the fatty acid composition or IV of one depot to predict the IV of another depot.

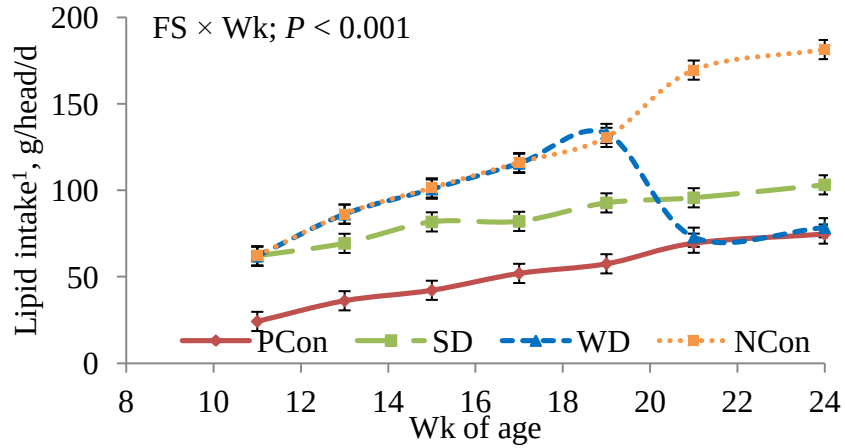
While generic acceptability thresholds for pork fat IV have been established, the economic value of a unit of change in IV has not been established. In general, the results of this study showed that carcass fat depots responded similarly among dietary feeding strategies using 2 different IV equations. However, IV of jowl, belly, and backfat using the Meadus equation was consistently greater than the IV using the AOCS equation. This occurred due to the inclusion of longer, more unsaturated fatty acids in the Meadus equation resulting in inconsistent changes in IV-diff among jowl, belly, and backfat depots and DDGS feeding strategies. Longer chain, more unsaturated fatty acids are

present in small quantities (e.g. eicosadienoic, homo-α-linolenic, arachidonic, docosahexaenoic), in pork jowl, belly, and backfat, but longer, more unsaturated fatty acids have lower melting points and are more likely to be peroxidized (Erickson, 2008). Given the lower melting point and peroxidation susceptibility of these longer, more unsaturated fatty acids, use of the AOCS IV equation as a measure of pork fat quality may be less accurate than using the Meadus IV equation.

Figure 6.1. Lipid intake of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies from 8 to 24 wk of age (See also Appendix Tables A.20 and A.21)



Time interval between 2nd Improvest® dose and harvest (TD)[2]	Wk of age						
	11	13	15	17	19	21	24
TD9					a	a	a
TD7					b	a	ab
TD5					b	b	b
TD9	A	B	C	D	EY	FX	F
TD7	A	B	C	C	D	D	D
TD5	A	B	C	C	D	D	D

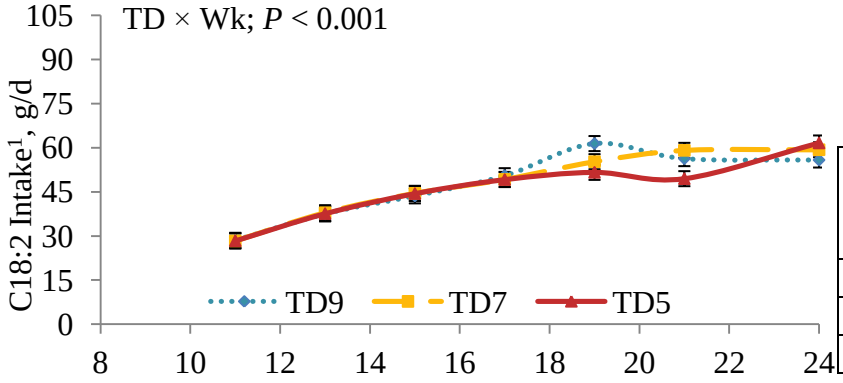


FS[3]	Wk of age						
	11	13	15	17	19	21	24
PCon	a	a	a	a	a	a	a
SD	b	b	b	b	b	b	b
WD	b	c	c	c	c	a	a
NCon	b	c	c	c	c	c	c
PCon	A	B	BC	CD	D	E	E
SD	A	A	BX	BX	CY	C	C
WD	AX	C	D	E	F	BY	BC
NCon	A	B	C	D	E	F	G

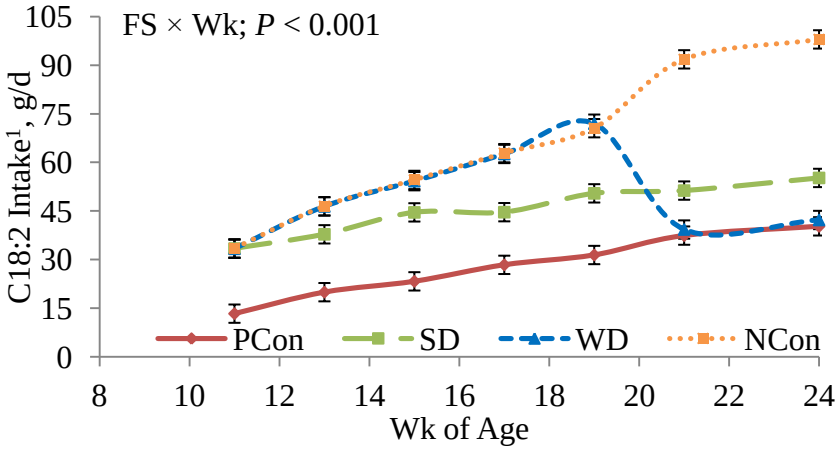
[1] Lipid intake was calculated as (total feed intake × analyzed dietary lipid content)/(pigs per pen × d on feed).
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[a,b,c] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a column, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F,G] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Figure 6.2. Linoleic acid (C18:2n-6) intake of immunologically castrated pigs harvested at 5, 7, or 9 weeks after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies during 8 to 24 wk of age (See also Appendix Tables A.20 and A.21)



Time interval between 2nd Improvest® dose and harvest (TD)[2]	Wk of age 11	13	15	17	19	21	24
TD9					a	a	a
TD7					b	a	ab
TD5					b	b	b
TD9	A	B	C	D	F	E	E
TD7	A	B	CX	CY	D	D	D
TD5	A	B	CX	CY	D	D	E



FS[3]	Wk of age 11	13	15	17	19	21	24
PCon	a	a	a	a	a	a	a
SD	b	bx	b	b	b	b	b
WD	b	cy	c	c	c	a	a
NCon	b	c	c	c	c	c	c
PCon	A	B	BC	CD	D	E	E
SD	A	A	BX	BX	CY	C	C
WD	A	C	D	E	F	B	BC
NCon	A	B	C	D	E	F	G

[1] C18:2n6 intake calculated as (total feed intake × analyzed dietary lipid content × analyzed dietary linoleic acid content)/ (pigs per pen × d on feed).

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[a,b,c] Within a column, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a column, means without a common superscript differ ($P \leq 0.10$).
[A,B,C,D,E,F,G] Within a row, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a row, means without a common superscript differ ($P \leq 0.10$).

Table 6.1. Ether extract and linoleic acid intake of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Tables A.19)

	Feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest(TD)[2]				*P* value		
Trait	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD	FS × TD
Ether extract intake[3], g/head/d	50.9[a]	83.9[bx]	92.7[by]	121.1[c]	5.2	88.1[a]	88.1[a]	85.1[b]	4.8	< 0.01	0.04	0.08
C18:2n6 intake[4], g/head/d	27.7[a]	45.4[b]	50.0[b]	65.4[c]	2.6	47.7[a]	47.7[a]	46.0[b]	2.4	< 0.01	0.03	0.07

[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] Ether extract intake calculated as (total feed intake × analyzed dietary ether extract)/(pigs per pen × d on feed).

[4] C18:2n6 intake calculated as (total feed intake × analyzed dietary ether extract × analyzed dietary linoleic acid content)/(pigs per pen × d on feed).

[a,b,c] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.10$).

Table 6.2. Morphometric belly characteristics of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Table A.22)

	Feeding strategy (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value[3]	
Item	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD
HCW, kg	90.6^{ax}	88.3^{by}	87.8^{by}	85.3^{b}	1.5	87.8	88.1	88.4	1.5	< 0.01	0.80
Belly (IMPS #408), % chilled side wt	13.56^{a}	13.07^{ab}	12.87^{ab}	12.20^{b}	0.95	13.10	12.71	12.97	0.93	0.04	0.62
Belly thickness[4], cm	2.97^{a}	2.95^{ax}	2.98^{a}	2.80^{by}	0.20	2.99^{x}	2.95	2.86^{y}	0.76	0.02	0.06
Belly width[4], cm	22.6^{a}	23.2^{ab}	23.0^{ab}	23.8^{b}	0.7	23.0	23.4	23.1	0.7	0.03	0.61
Belly length[4], cm	61.7	61.4	61.6	61.3	1.5	61.3	60.9^{x}	62.3^{y}	1.4	0.94	0.06
Flop distance[4], cm	8.66^{a}	7.34^{b}	7.11^{b}	5.57^{c}	0.65	7.26	7.47	6.79	0.62	< 0.01	0.24
Flop angle[5,6], °	16.4^{ax}	14.0^{by}	13.4^{b}	10.3^{c}	1.4	13.9	14.1	12.6	1.4	< 0.01	0.16

[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] FS × TD ($P \geq 0.27$).

[4] HCW was used as a covariate ($P < 0.01$).

[5] HCW was not used as a covariate ($P > 0.05$).

[6] Belly flop angle calculated as: $\cos^{-1}[0.5(L^2) - D^2]/[0.5(L^2)]$, where L = belly length,cm and d = skin-side down, skin-to-skin belly distance,cm (Whitney et al., 2006).

[a,b,c] Least squares means with different superscripts differ ($P < 0.05$).

[x,y] Least squares means with different superscripts differ ($P < 0.10$).

Table 6.3. Objective and subjective adipose color from 3 depot locations when immunologically castrated pigs were harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies (see also Appendix Table A.23).

	Feeding strategies (FS)[1]					Interval between second Improvest® dose and harvest (TD)[2]				*P* value[3]	
Item	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	FS	TD
Jowl adipose[4]											
L*	74.8	75.1	75.5	74.7	1.7	75.5^{ax}	73.9^{by}	75.7^{a}	1.6	0.79	0.03
a*	0.47	0.36	0.60	0.09	0.20	0.35	0.49	0.29	0.17	0.27	0.67
b*	5.93	6.33	6.65	6.28	0.65	5.92	6.59	6.39	0.60	0.74	0.47
Back adipose[4]											
L*	76.5	76.4	76.8	75.2	1.6	76.4	76.4	75.9	1.5	0.17	0.65
a*	0.50	0.30	0.70	0.17	0.28	0.38	0.53	0.34	0.27	0.21	0.68
b*	6.65	6.25	6.98	6.55	0.73	6.59	6.59	6.63	0.72	0.43	0.99
Belly adipose[4]											
L*	78.6^{a}	78.1^{a}	77.6^{ab}	76.5^{b}	0.6	77.7	77.7	77.7	0.6	< 0.01	1.00
a*	-0.02	-0.07	0.14	-0.25	0.20	-0.11	0.12	-0.16	0.20	0.19	0.14
b*	5.46	5.04	5.78	5.41	0.46	5.33	5.63	5.31	0.43	0.38	0.62
JCS[5]	1.08^{ax}	1.15^{a}	1.29^{aby}	1.33^{b}	0.07	1.16	1.20	1.28	0.07	0.02	0.26

[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] FS × TD ($P \geq 0.33$).

[4] Objective Hunter color values L* (100 = white, 0 = black), a* (positive = more red, negative = more green), b* (positive = more yellow, negative = more blue).

[5] JCS = Japanese Color Score (1 = white and 4 = yellow).

[a,b] Least squares means with different superscripts differ ($P < 0.05$).

[x,y] Least squares means with different superscripts differ ($P < 0.10$).

Table 6.4. Fatty acid composition and calculated iodine value (IV) of 3 adipose depots from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies (FS; see also Appendix Table A.24)[1]

	Jowl				Back				Belly					*P* value
Item	PCon	SD	WD	NCon	PCon	SD	WD	NCon	PCon	SD	WD	NCon	SEM	FS × Depot
Crude Fat[2], %	88.7	88.4	88.6	88.0	88.0	88.9	88.4	88.3	88.4	88.3	87.9	88.7		0.04
SFA														
C14:0 – Myristic	1.40	1.28	1.22	1.17	1.42	1.33	1.31	1.20	1.45	1.33	1.28	1.21	0.02	0.37
C16:0 – Palmitic	23.5^{c}	22.1^{ey}	21.7^{ef}	20.4^{g}	25.9^{a}	24.6^{b}	25.2^{ab}	22.4^{de}	24.5^{b}	23.2^{cd}	23.0^{cdx}	21.1^{fg}	0.3	< 0.01
C18:0 – Stearic	10.5^{cd}	9.8^{de}	9.7^{de}	8.6^{f}	14.2^{a}	13.0^{b}	13.8^{ab}	11.0^{c}	11.1^{c}	10.5^{cd}	10.4^{cd}	8.9^{ef}	0.3	< 0.01
C20:0 – Arachidic	0.234	0.233	0.227	0.220	0.274	0.268	0.270	0.262	0.250	0.247	0.250	0.238	0.007	0.99
MUFA														
C18:1n9 - Oleic	39.0^{b}	37.2^{cd}	37.0^{cd}	36.1^{dey}	37.4^{cx}	35.4^{e}	35.5^{e}	32.9^{f}	40.2^{a}	37.8^{bc}	37.4^{cx}	35.4^{e}	0.5	< 0.01
C20:1n9 - Gonodic	0.92^{ax}	0.86^{abc}	0.84^{bcy}	0.85^{abc}	0.84^{bc}	0.85^{abc}	0.80^{c}	0.83^{bc}	0.86^{abc}	0.85^{abc}	0.89^{ab}	0.85^{abc}	0.02	0.02
PUFA														
C18:2n6 - Linoleic	13.2^{f}	18.1^{bc}	19.4^{b}	23.4^{a}	10.6^{g}	15.7^{de}	14.7^{ef}	22.7^{a}	11.4^{g}	16.3^{de}	17.1^{cd}	22.9^{a}	0.9	< 0.01
C18:3n3 - Linolenic	0.73^{cdex}	0.78^{bcd}	0.81^{b}	0.91^{a}	0.60^{g}	0.64^{fgy}	0.62^{g}	0.80^{bc}	0.60^{g}	0.64^{fg}	0.71^{def}	0.79^{bcd}	0.04	0.04
C20:2 - Eicosadienoic	0.74^{f}	0.89^{bcde}	0.91^{bc}	1.12^{a}	0.69^{f}	0.77^{f}	0.71^{f}	0.98^{b}	0.78^{efy}	0.80^{cdef}	0.90^{bcdx}	0.98^{b}	0.04	< 0.01
C20:4n6 - Arachidonic	0.40^{cde}	0.45^{abcx}	0.467^{b}	0.50^{a}	0.34^{e}	0.37^{de}	0.34^{e}	0.36^{de}	0.38^{de}	0.39^{dey}	0.38^{de}	0.43^{bcd}	0.02	0.06
Total SFA[3]	36.6^{cd}	34.2^{ef}	33.8^{fg}	31.3^{h}	42.7^{a}	40.0^{b}	41.3^{ab}	35.7^{de}	38.1^{c}	36.2^{d}	35.8^{de}	32.4^{gh}	0.5	< 0.01
Total MUFA[4]	45.3^{ab}	42.9^{bcd}	43.5^{bcd}	42.2^{cd}	43.0^{bcd}	41.6^{d}	41.7^{dy}	38.6^{e}	47.5^{a}	44.3^{bcx}	44.1^{bcx}	41.4^{d}	0.7	0.07
Total PUFA[5]	15.3^{gm}	20.4^{c}	21.9^{c}	26.2^{ax}	12.1^{h}	17.2^{efkl}	16.2^{fg}	24.5^{by}	13.3^{h}	18.4^{de}	19.2^{dj}	25.3^{ab}	1.0	< 0.01
Total n3[6]	0.90^{cd}	0.97^{bc}	1.01^{b}	1.12^{a}	0.74^{ef}	0.80^{dex}	0.76^{ef}	0.96^{bc}	0.71^{fy}	0.76^{ef}	0.83^{de}	0.90^{cd}	0.05	0.05
Total n6[7]	13.6^{f}	18.5^{bc}	19.9^{b}	23.9^{a}	11.0^{g}	16.1^{de}	15.1^{ef}	23.1^{a}	11.8^{g}	16.8^{de}	17.5^{cd}	23.3^{a}	0.9	< 0.01
n6:n3	15.6^{fg}	19.3^{eyjl}	19.8^{de}	21.4^{cdx}	15.2^{gl}	20.3^{de}	20.2^{de}	24.3^{b}	17.2^{fk}	22.6^{bc}	21.4^{cdx}	26.6^{a}	0.5	< 0.01
IV-AOCS[8]	61.9^{f}	68.5^{cd}	70.6^{bc}	76.7^{a}	55.3^{g}	62.1^{f}	60.5^{f}	72.1^{b}	59.4^{f}	65.5^{e}	66.7^{de}	74.9^{a}	1.3	< 0.01
IV-Meadus[8]	64.6^{f}	71.5^{cd}	73.8^{bc}	80.3^{a}	57.8^{g}	64.9^{f}	63.1^{f}	75.2^{b}	62.1^{f}	68.3^{e}	69.6^{de}	78.0^{a}	1.3	< 0.01
IV difference	2.68^{d}	3.05^{bc}	3.16^{b}	3.59^{a}	2.57^{d}	2.81^{cd}	2.63^{d}	3.08^{bc}	2.69^{d}	2.77^{cd}	2.81^{cd}	3.14^{b}	0.11	< 0.01

[1]PCon = pigs fed corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in 4 dietary phases, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.
[2] For crude fat, with Tukey adjustment Diet × Depot comparisons were not significantly different ($P > 0.05$).
[3] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.
[4] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10 Margaroleic] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].
[5] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2 Eicosadienoic] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].
[6]Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].
[7] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].
[8]Iodine Value (IV) - AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV – Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930) (Meadus et al., 2010).
[9] IV differences = (IV-AOCS) – (IV-Meadus).
[a,b,c,d,e,f,g] Within a row, means without a common superscript differ ($P < 0.05$).
[x,y] Within a row, means without a common superscript differ ($P < 0.10$).
[j,k] Within a row, means without a common superscript differ ($P < 0.10$).
[l ,m] Within a row, means without a common superscript differ ($P < 0.10$).

Table 6.5. Main effects of harvesting immunologically castrated pigs at 5, 7, or 9 wk after the second Improvest® and feeding corn dried distillers grains with solubles (DDGS) strategies, on fatty acid composition and calculated Iodine Value (IV) of 3 adipose depot locations (See also Appendix A.25)

	DDGS Feeding Strategy (FS)[1]					Interval between 2nd Improvest® dose and harvest (TD)[2]				Fat Depot				*P* value[3,4,5]		
Item	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	Jowl	Back	Belly	SEM	FS	TD	Depot
Crude Fat, %	88.36	88.55	88.31	88.35	0.18	88.59	88.31	88.27	0.15	88.43	88.34	88.41	0.14	0.78	0.28	0.88
SFA																
C14:0 – Myristic	1.422^{a}	1.31^{b}	1.27^{b}	1.19^{c}	0.02	1.31	1.31	1.28	0.02	1.27^{a}	1.32^{b}	1.32^{b}	0.02	< 0.01	0.19	< 0.01
C16:0 – Palmitic	24.64^{a}	23.3^{b}	23.31^{b}	21.32^{c}	0.24	23.25^{a}	23.30^{ax}	22.88^{y}	0.23	21.94^{a}	24.54^{b}	22.95^{c}	0.21	< 0.01	0.05	< 0.01
C18:0 – Stearic	11.93^{a}	11.08^{b}	11.29^{b}	9.51^{c}	0.20	11.10	11.00	10.76	0.19	9.65^{a}	12.97^{b}	10.25^{c}	0.17	< 0.01	0.19	< 0.01
C20:0 – Arachidic	0.253	0.249	0.249	0.240	0.005	0.249	0.248	0.246	0.004	0.228^{a}	0.269^{b}	0.246^{c}	0.004	0.23	0.91	< 0.01
MUFA																
C18:1n9 - Oleic	38.85^{a}	36.83^{b}	36.64^{b}	34.8^{c}	0.49	36.9	36.94	36.49	0.48	37.34^{ax}	35.29^{b}	37.71^{ay}	0.46	< 0.01	0.17	< 0.01
C20:1n9 - Gonodic	0.87	0.85	0.84	0.84	0.02	0.85	0.86	0.85	0.01	0.87^{a}	0.83^{b}	0.86^{a}	0.01	0.13	0.73	< 0.01
PUFA																
C18:2n6 - Linoleic	11.73^{a}	16.68^{b}	17.08^{b}	22.97^{c}	0.86	16.7^{a}	16.81^{a}	17.83^{b}	0.84	18.51^{a}	15.92^{c}	16.91^{b}	0.83	< 0.01	< 0.01	< 0.01
C18:3n3 - Linolenic	0.64^{a}	0.69^{b}	0.71^{b}	0.83c	0.04	0.71^{a}	0.70^{a}	0.75^{b}	0.04	0.81^{a}	0.66^{b}	0.68^{b}	0.04	< 0.01	< 0.01	< 0.01
C20:2 - Eicosadienoic	0.74^{a}	0.82^{b}	0.84^{b}	1.02^{c}	0.03	0.86	0.85	0.86	0.03	0.91^{a}	0.79^{c}	0.86^{b}	0.03	< 0.01	0.75	< 0.01
C20:4n6 - Arachidonic	0.37^{a}	0.40^{bc}	0.39^{ab}	0.43^{c}	0.02	0.39^{x}	0.40	0.41^{y}	0.01	0.45^{a}	0.35^{c}	0.39^{b}	0.01	< 0.01	0.08	< 0.01
Total SFA[6]	39.1^{a}	36.8^{b}	36.9^{b}	33.1^{c}	0.4	36.8^{x}	36.7	36.0^{y}	0.4	34.0^{a}	39.9^{c}	35.6^{b}	0.4	< 0.01	0.07	< 0.01
Total MUFA[7]	45.3^{a}	42.9^{b}	43.1^{b}	40.8^{c}	0.6	42.9	43.2	43.0	0.6	43.5^{ax}	41.2^{b}	44.3^{ay}	0.5	< 0.01	0.73	< 0.01
Total PUFA[8]	13.6^{a}	18.7^{b}	19.1^{b}	25.3^{c}	0.9	18.8^{a}	18.8^{a}	19.9^{b}	0.9	21.0^{a}	17.5^{c}	19.0^{b}	0.9	< 0.01	< 0.01	< 0.01
Total n3[9]	0.78^{a}	0.84^{b}	0.87^{b}	0.99^{c}	0.05	0.86^{a}	0.85^{a}	0.90^{b}	0.05	1.00^{a}	0.82^{b}	0.80^{b}	0.05	< 0.01	0.02	< 0.01
Total n6[10]	12.1^{a}	17.1^{b}	17.5^{b}	23.4^{c}	0.9	17.1^{a}	17.2^{a}	18.3^{b}	0.9	19.0^{a}	16.3^{c}	17.3^{b}	0.8	< 0.01	< 0.01	< 0.01
n6:n3	16.0	20.7	20.5	24.1	0.3	20.2	20.4	20.5	0.3	19.0	21.9	20.0	0.2	< 0.01	0.67	< 0.01
IV-AOCS[11]	58.9^{a}	65.4^{b}	65.9^{b}	74.6^{c}	1.2	65.5^{a}	65.8^{a}	67.2^{b}	1.2	69.4^{a}	66.6^{b}	62.5^{c}	1.2	< 0.01	< 0.01	< 0.01
IV-Meadus[11]	61.5^{a}	68.2^{b}	68.8^{b}	77.8^{c}	1.3	68.4^{a}	68.7^{a}	70.2^{b}	1.3	72.5^{a}	69.5^{b}	65.2^{c}	1.3	< 0.01	< 0.01	< 0.01
IV difference[12]	2.65^{a}	2.88^{b}	2.86^{b}	3.27^{c}	0.09	2.89	2.89	2.97	0.09	3.12^{a}	2.77^{b}	2.85^{b}	0.09	< 0.01	0.26	< 0.01

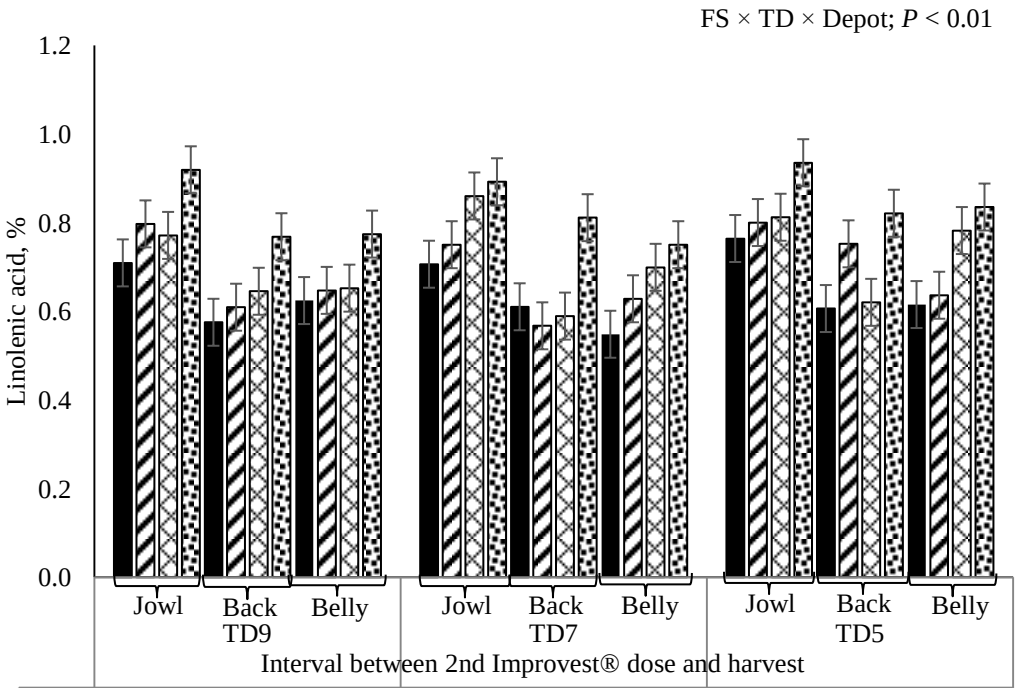
[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[3] FS × TD × depot; ($P > 0.05$) except for C18:1n-9 - Oleic, C18:3n-3 - linolenic, Total n3, and n3:n6 (See Appendix, Figure A.2, Chapter 6, Figure 6.3, Appendix Figures A.6, A.7, respectively).
[4]TD × depot; ($P > 0.05$) except for C23:0 Tricosylic, C15:1n-10 Pentadecenoic, C20:3n3 - Homo-α-linolenic, C20:3n6 - Homo-γ-linolenic, C22:5n3 - Clupanodonic, n3:n6 (See Appendix Figures A.1, A.3, A.5, A.5, A.5, A.8, respectively and Appendix Table A.27).
[5]FS × TD; ($P > 0.05$) except for C10:0 Capric, C14:1n-9 - Myristoleic, C18:1n9t - Elaidic, C20:3n3 - Homo-α-linolenic, C20:3n6 - Homo-γ-linolenic (See Appendix Figures A.1, A.3, A.3, A.4, A.4, respectively and Appendix Table A.26).
[6] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.
[7] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10 Margaroleic] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].
[8] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2 Eicosadienoic] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].
[9] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].
[10] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].
[11] Iodine Value (IV) - AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV – Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930; Meadus et al., 2010).
[12] IV differences = (IV-AOCS) – (IV-Meadus).
[a,b,c] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.05$).
[x,y] Within a row of a single main effect, means without a common superscript differ ($P \leq 0.10$).

Table 6.6. Comparison of fatty acids coefficients used in two different iodine value (IV) equations

Fatty acid	IV-AOCS[1]	IV-Meadus[2]
C16:1c	0.950	0.950
C18:1n-9	0.860	0.860
C18:2n-6	1.732	1.732
C18:3n-3	2.616	2.616
C20:1n-9	0.785	0.795
C20:2	--	1.570
C20:3n-3	--	2.380
C20:4n-6	--	3.190
C20:5n-3	--	4.010
C22:1n-9	0.723	--
C22:4n-6	--	2.930
C22:5n-3	--	3.680
C22:6n-3	--	2.930

[1] (AOCS, 1998)
[2] (Meadus et al., 2010)

Figure 6.3. Interactive effect of harvesting immunologically castrated pigs at 5, 7, or 9 wk after the second Improvest® dose and feeding corn dried distillers grains with solubles (DDGS) feeding strategies (FS) on linolenic acid content of 3 adipose depots[1,2]



		TD9	TD7	TD5
Jowl	PCon	0.709	0.706	0.764
	SD	0.797	0.750	0.800
	WD	0.771	0.860	0.812
	NCon	0.919	0.892	0.935
Back	PCon	0.575	0.610	0.606
	SD	0.609	0.567	0.752
	WD	0.645	0.589	0.620
	NCon	0.768	0.811	0.821
Belly	PCon	0.624	0.548	0.615
	SD	0.647	0.628	0.636
	WD	0.652	0.699	0.782
	NCon	0.774	0.750	0.835

[1]PCon = pigs fed corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in 4 dietary phases, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

CHAPTER 7: SUMMARY

In pork production, feed cost represents the largest proportion of total production costs and energy represents the greatest component of feed cost (Schulz, 2014; Woyengo et al., 2014). Thus, great emphasis is placed on identifying feed ingredients to supply pigs with the necessary dietary calories and nutrients, and to improve nutrient utilization and lean gain efficiency, to subsequently optimize lean muscle accretion and reduce the cost of lean growth.

Intact male (**IM**) pigs have reduced feed intake, carcass fat, and improved lean gain efficiency compared to physical castrates (**PC**), but PC are castrated at an early age to avoid unpalatable off-odors known as boar taint (Squires, 2011). In 2011, the U.S. Food and Drug Administration approved immunological castration of IM pigs using Improvest®, with the second Improvest® dose administered 3 to 10 weeks before harvest to minimize boar taint (FDA, 2011a). Immunological castration captures the lean gain efficiency of IM pigs while minimizing boar taint in fresh and processed pork products. However, immunologically castrated (**IC**) pigs typically have lower carcass dressing percentage (Lawrence et al., 2012) compared with PC due to greater visceral mass (Boler et al., 2014). As the time interval between the second Improvest® dose and harvest increases from 3 to 10 weeks, ADFI and carcass fat of IC pigs increases (Lealiifano et al., 2011; Elsbernd et al., 2014);. Fat deposition requires more dietary energy than lean deposition (Noblet and van Milgen, 2013). Thus, reducing carcass fat and increasing carcass lean reduces the cost of pork production. However, pigs with less backfat have softer pork fat (Wood et al., 1989). Soft pork fat can create challenges in handling and

processing for packers, and undesirable appearance and flavors for consumers (Wood et al., 2008).

Soft pork fat is characterized by high concentrations of polyunsaturated fatty acids in adipose tissue, and is caused by feeding diets containing ingredients with high concentrations of polyunsaturated fatty acids (Apple et al., 2009b) to gilts and PC. Corn dried distillers grains with solubles (**DDGS**) has become an attractive feed ingredient for growing-finishing swine diets due to its affordability and desirable energy, protein, lipid and phosphorus content (Stein and Shurson, 2009). Feeding diets containing up to 40% (Graham et al., 2014) and 60% (Leick et al., 2010; Hardman, 2014) DDGS can reduce carcass dressing percentage. Thus, the cost of production per unit of carcass weight increases when feeding DDGS diets. The reduction in carcass dressing percentage has been attributed to increased visceral mass and/or gut fill due to feeding high fiber ingredients (Agyekum et al., 2012; Asmus et al., 2014a). The lipid content of DDGS is high in polyunsaturated fatty acids (NRC, 2012). Therefore, feeding increasing dietary levels of DDGS can result in softer pork fat in PC and gilts (Xu et al., 2010b; McClelland et al., 2012). Due to the decreased carcass fat of IC pigs compared to PC pigs (Boler et al., 2012; Tavárez et al., 2014b), IC pigs may also have softer pork fat when fed increasing dietary levels of DDGS. To overcome the negative impact of feeding DDGS diets to gilts and PC pigs on pork fat quality, DDGS can be removed from the diet for several weeks prior to harvest to improve pork fat firmness (Xu et al., 2010a) and carcass dressing percentage (Gaines et al., 2007). Additionally, pork producers have adopted the use of other feeding strategies such as gradually reducing dietary DDGS inclusion during

the growing-finishing period (**SD**) to overcome soft pork fat and reduced carcass dressing percentage, but these strategies have not been experimentally evaluated.

Therefore, one of the overall objectives of the research represented in this thesis was to evaluate the effects of using 4 DDGS feeding strategies for IC pigs on carcass dressing percentage and pork fat firmness. A second overall objective was to evaluate the effects of these feeding strategies for IC pigs that are harvested at 5 (**TD5**), 7 (**TD7**), or 9 (**TD9**) weeks after the second Improvest® dose.

In chapter 2, pigs fed diets with 40% DDGS (**NCon**) had reduced ADFI compared with pigs fed a corn-soybean meal control diet (**PCon**), which may have been due to limited gut capacity when the high fiber DDGS diets were fed. Interestingly, when 40% DDGS was withdrawn from the diet for 5 weeks before harvest (**WD**), ADFI increased so that during the 21 to 24 week interval, pigs fed WD had greater ADFI compared with pigs fed PCon, SD, and NCon. Pigs harvested 9 weeks after the second Improvest® dose appeared to be more affected by DDGS feeding strategies compared with TD5 and TD7 pigs, because TD9 pigs fed PCon and WD had greater overall ADFI than TD9 pigs fed NCon. Overall ADFI was not different among dietary feeding strategies for TD5 or TD7 pigs. The increases in ADFI after the second Improvest® dose also resulted in greater ME intake as discussed in chapter 3. It appears that the increased ME intake that occurred when DDGS was removed from the diet was deposited as lean and fat, as determined by live animal ultrasound evaluation of the LM and backfat in chapter 4.

Serum IGF-1 and leptin were evaluated in chapter 4 as biological markers of growth. However, changes in serum concentrations of IGF-1 and leptin due to feeding strategy and timing of the second Improvest dose were inconsistent with live animal

ultrasound of the LM and backfat. The increase in ADFI once DDGS was removed from the diet at 19 WOA coincided with pigs fed WD having greater backfat and serum leptin concentrations than pigs fed NCon at 21 and 24 WOA in TD9 pigs. However, an increase in serum leptin concentrations should have resulted in reduced ADFI. Thus, other factors may be modulating the relationship among ADFI, backfat thickness, and serum leptin concentrations.

Pigs fed SD and PCon had similar body composition and ADFI. Thus, lean gain per day and lean gain efficiency was also similar between pigs fed SD and PCon. However, these similarities in body composition, ADFI, lean gain per day and lean gain efficiency relative to pigs fed PCon were not achieved by using the WD strategy. Additionally, the SD diets were less expensive than PCon diets, thus the SD feeding strategy was more advantageous at reducing the diet cost of lean gain than the WD feeding strategy. In fact, diet cost of lean gain when using the SD strategy was lower than using the PCon strategy.

Chapters 5 and 6 evaluated pork loin quality and pork fat quality, respectively. Feeding strategy had a much greater impact on pork loin and belly quality, and fatty acid composition of jowl, belly, and backfat than the time interval between the second Improvest® dose and harvest. Feeding diets with 40% DDGS resulted in reduced ham, loin, and belly primal cut yields and softer pork loins with less marbling compared with pigs fed corn-soybean meal diets. Withdrawing 40% DDGS from the diets 5 weeks before harvest improved marbling, but not loin firmness, while gradually decreasing the DDGS throughout the growing-finishing period improved loin firmness but not marbling. Both of the SD and WD feeding strategies increased the concentration of saturated fatty

acids in jowl, belly, and backfat compared with pigs fed NCon. Pork loin quality was unaffected by the timing between the second Improvest® dose and harvest. Bellies from TD9 pigs tended to be thicker than bellies from TD5 pigs. Increasing the time interval between the second Improvest® dose and harvest from 5 to 7 weeks increased the saturated fatty acid content of jowl, belly, and backfat, indicating firmer pork fat. A commonly used composite assessment of pork fat quality is the calculation of iodine value (**IV**) where larger coefficients are placed on longer, more unsaturated fatty acids resulting in higher IV and indicating undesirable soft pork fat. In this study, IV of jowl, belly, and backfat of all TD treatments, regardless of IV equation, were lower than commonly used acceptability IV thresholds that range from 70 (Barton-Gade, 1987) to 75 (Boyd et al., 1997).

However, multiple IV equations have been developed (AOCS, 1998; Meadus et al., 2010) but are different because the Meadus IV equation includes additional fatty acids that are longer and more unsaturated compared with the AOCS IV equation. Iodine value of jowl, belly, and backfat was greater when using the Meadus IV equation compared with the AOCS IV equation. However, the IV difference between the Meadus and AOCS equation was not consistent among fat depots and DDGS feeding strategies.

In conclusion, regulation of feed intake due to DDGS feeding strategy and the timing of the second Improvest® dose need to be further refined to formulate diets with appropriate nutrient concentrations to optimize economic and environmental sustainability of pork production when using these technologies. It is possible that pig age at the time when the second dose of Improvest® is administered, influences the rate of ADFI increase. Feeding diets containing 40% DDGS to IC pigs reduces ADFI,

particularly when pigs are harvested 9 weeks after the second Improvest® dose. Additionally, all IC pigs fed diets containing 40% DDGS had reduced carcass fat thickness, lower primal cut yields, and softer more unsaturated, jowl, belly, and backfat compared with pigs fed corn-soybean meal diets. The SD feeding strategy was more effective than the WD feeding strategy in maintaining similar lean gain per day and lean gain efficiency to pigs fed PCon.

In recent years, concerns regarding pork fat quality have increased because of the widespread use of DDGS in growing-finishing diets in North America. The lean quality responses using these DDGS feeding strategies for IC pigs observed in this study, suggest that more emphasis should also to be placed on pork loin quality. This is the first study to compare IV equations relative to assessing pork fat quality. Given the lower melting point and greater susceptibility of longer, more unsaturated fatty acids to become peroxidized, the Meadus IV equation may provide more accurate distinction between acceptable and unacceptable pork fat quality.

Literature Cited

Aberle, E. D., J. C. Forrest, D. E. Gerrard, and E. W. Mills. 2001. Principles of Meat Science. 4th ed. Kendall/Hunt Publishing Company, Dubuque.

Agyekum, A. K., B. A. Slominski, and C. M. Nyachoti. 2012. Organ weight, intestinal morphology, and fasting whole-body oxygen consumption in growing pigs fed diets containing distillers dried grains with solubles alone or in combination with a multienzyme supplement. J. Anim. Sci. 90: 3032-3040.

Alexandratos, N., and J. Bruinsma. 2012. World agriculture towards 2030/2050: the 2012 revision, FAO, Rome.

Almeida, F. N., G. I. Petersen, and H. H. Stein. 2011. Digestibility of amino acids in corn, corn coproducts, and bakery meal fed to growing pigs. J. Anim. Sci. 89: 4109-4115.

Almeida, F. N., and H. H. Stein. 2010. Performance and phosphorus balance of pigs fed diets formulated on the basis of values for standardized total tract digestibility of phosphorus. J. Anim. Sci. 88: 2968-2977.

Aluwé, M., S. Millet, K. M. Bekaert, F. A. M. Tuyttens, L. Vanhaecke, S. De Smet, and D. L. De Brabander. 2011. Influence of breed and slaughter weight on boar taint prevalence in entire male pigs. Animal 5: 1283-1289.

Ambrosen, K. 1993. Dry feeding strategy and fasting periode for entire male pigs, Pig Research Centre, Copenhagen, Denmark.

Ames, J. M. 1998. Applications of the Maillard reaction in the food industry. Food Chem. 62: 431-439.

Anderson, P. V., B. J. Kerr, T. E. Weber, C. J. Ziemer, and G. C. Shurson. 2012. Determination and prediction of digestible and metabolizable energy from chemical analysis of corn coproducts fed to finishing pigs. J. Anim. Sci. 90: 1242-1254.

Andersson, K., A. Schaub, K. Andersson, K. Lundström, S. Thomke, and I. Hansson. 1997. The effects of feeding system, lysine level and gilt contact on performance, skatole levels and economy of entire male pigs. Livest. Prod. Sci. 51: 131-140.

Andresen, O. 2006. Boar taint related compounds: Androstenone/skatole/other substances. Acta Agric. Scand. 48: S5-S8.

AOCS. 1998. Official Methods and Recommended Practices of the AOCS. 5th ed. Sm. Oil Chem. Soc., Champaign, IL.

AOCS. 2009. Arthur von Hübl and the iodine value. http://lipidlibrary.aocs.org/history/Hubl/index.htm Accessed October 28, 2014.

Apple, J. K. 2010. Beyond Iodine Value "Measuring Belly/Fat Firmness" Reciprocal Meat Conference, Lubbock, TX.

Apple, J. K., C. V. Maxwell, D. C. Brown, K. G. Friesen, R. E. Musser, Z. B. Johnson, and T. A. Armstrong. 2004. Effects of dietary lysine and energy density on performance and carcass characterisitics of finishing pigs fed ractopamine. J. Anim. Sci. 82: 3277-3287.

Apple, J. K., C. V. Maxwell, D. L. Galloway, C. R. Hamilton, and J. W. S. Yancey. 2009a. Interactive effects of dietary fat source and slaughter weight in growing-

finishing swine: II. Fatty acid composition of subcutaneous fat. J. Anim. Sci. 87: 1423-1440.

Apple, J. K., C. V. Maxwell, D. L. Galloway, C. R. Hamilton, and J. W. S. Yancey. 2009b. Interactive effects of dietary fat source and slaughter weight in growing-finishing swine: III. Carcass and fatty acid compositions. J. Anim. Sci. 87: 1441-1454.

Apple, J. K., C. V. Maxwell, D. L. Galloway, S. Hutchison, and C. R. Hamilton. 2009c. Interactive effects of dietary fat source and slaughter weigh in growing-finishing swine. I. Growth performance and longissimus muscle fatty acid composition. J. Anim. Sci. 87: 1407-1422.

Apple, J. K., C. V. Maxwell, J. T. Sawyer, B. R. Kutz, L. K. Rakes, M. E. Davis, Z. B. Johnson, S. N. Carr, and T. A. Armstrong. 2007. Interactive effect of ractopamine and dietary fat source on quality characterisitics of fresh pork bellies. J. Anim. Sci. 85: 2682-2690.

Apple, J. K., J. T. Sawyer, C. V. Maxwell, J. W. S. Yancey, J. W. Frank, J. C. Woodworth, and R. E. Musser. 2011. Effects of l-carnitine supplementation on quality characteristics of fresh pork bellies from pigs fed 3 levels of corn oil. J. Anim. Sci. 89: 2878-2891.

Asmus, M. D., J. M. DeRouchey, M. D. Tokach, S. S. Dritz, T. A. Houser, J. L. Nelssen, and R. D. Goodband. 2014a. Effects of lowering dietary fiber before marketing on finishing pig growth performance, carcass characteristics, carcass fat quality, and intestinal weights. J. Anim. Sci. 92: 119-128.

Asmus, M. D., M. A. Tavarez, M. D. Tokach, S. S. Dritz, A. L. Schroeder, J. L. Nelssen, R. D. Goodband, and J. M. DeRouchey. 2014b. The effects of immunological castration and corn dried distillers grains with solubles withdrawal on growth performance, carcass characteristics, fatty acid analysis, and iodine value of pork fat depots. J. Anim. Sci. 92: 2116-2132.

Averette Gatlin, L., M. T. See, J. A. Hansen, D. Sutton, and J. Odle. 2002. The effects of dietary fat sources, levels, and feeding intervals on pork fatty acid composition. J. Anim. Sci. 80: 1606-1615.

Azain, M. J. 2001. Fat in Swine Nutrition. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. CRC Press, Boca Raton.

Babol, J., and E. J. Squires. 1995. Quality of meat from entire male pigs. Food Research International 28: 201-212.

Babol, J., E. J. Squires, and K. Lundstrom. 1999. Relationship between metabolism of androstenone and skatole in intact male pigs. J. Anim. Sci. 77: 84-92.

Badioo, S. K., J. Jendza, X. Yang, and L. Anil. 2014. Liquid feeding of ethanol industry co-products on growth performance of wean to finish pigs. J. Anim. Sci. 92:(Suppl. 2): 152(Abstr.).

Baker, D. H., and H. H. Stein. 2013. Bioavailability of minerals and vitamins in feedstuffs. In: L. I. Chiba (ed.) Sustainable Swine Nutrition. p 341-364. John Wiley & Sons, Inc., Ames.

Barb, C. R., G. J. Hausman, and K. L. Houseknect. 2001. Biology of leptin in the pig. Domest. Anim. Endocrinol. 21: 297-317.

Barbut, S., A. A. Sosnicki, S. M. Lonergan, T. Knapp, D. C. Ciobanu, L. J. Gatcliffe, E. Huff-Lonergan, and E. W. Wilson. 2008. Progress in reducing the pale, soft and exudative (PSE) problem in pork and poultry meat. Meat Sci. 79: 46-63.

Barton-Gade, P. A. 1987. Meat and fat quality in boars, castrates and gilts. Livest. Prod. Sci. 16: 187-196.

Batorek, N., M. Škrlep, A. Prunier, I. Louveau, J. Noblet, M. Bonneau, and M. Čandek-Potokar. 2012. Effect of feed restriction on hormones, performance, carcass traits, and meat quality in immunocastrated pigs. J. Anim. Sci. 90: 4593-4603.

Bee, G., S. Gebert, and R. Messikommer. 2002. Effect of dietary energy supply and fat source on the fatty acid pattern of adipose and lean tissues and lipogenesis in the pig. J. Anim. Sci. 80: 1564-1574.

Belyea, R., K. Rausch, T. Clevenger, V. Singh, D. Johnston, and M. Tumbleson. 2010. Sources of variation in composition of DDGS. Anim. Feed Sci. Technol. 159: 122-130.

Benz, J. M., S. K. Linneen, M. D. Tokach, S. S. Dritz, J. L. Nelssen, J. M. DeRouchey, R. D. Goodband, R. C. Sulabo, and K. J. Prusa. 2010. Effects of dried distillers grains with solubles on carcass fat quality of finishing pigs. J. Anim. Sci. 88: 3666-3682.

Benz, J. M., M. D. Tokach, S. S. Dritz, J. L. Nelssen, J. M. DeRouchey, R. C. Sulabo, and R. D. Goodband. 2011a. Effects of dietary iodine value product on growth performance and carcass fat quality of finishing pigs. J. Anim. Sci. 89: 1419-1428.

Benz, J. M., M. D. Tokach, S. S. Dritz, J. L. Nelssen, J. M. DeRouchey, R. C. Sulabo, and R. D. Goodband. 2011b. Effects of increasing choice white grease in corn- and sorghum-based diets on growth performance, carcass characterisitics, and fat quality characterisitcs of finishing pigs. J. Anim. Sci. 89: 773-782.

Berg, E. P., and S. J. Eilert. 1995. Critical points affecting fresh pork quality within the packing plant, National Pork Producers Council, Des Moines, IA.

Bergstrom, J. R., M. D. Tokach, S. S. Dritz, J. L. Nelssen, J. M. DeRouchey, and R. D. Goodband. 2009a. Effects of feeder design, gender, and dietary concentration of dried distillers grains with solubles on the growth performance and carcass characterisitcs of growing-finishing pigs Swine Day, Manhattan, KS.

Bergstrom, J. R., M. D. Tokach, S. S. Dritz, J. L. Nelssen, J. M. DeRouchey, and R. D. Goodband. 2009b. Effects of feeder design, gender, and dietary concentration of dried distillers grains with solubles on the growth performance and carcass characteristics of growing-finishing pigs, Kansas State University, Manhattan, KS.

Black, J. L., B. A. Williams, and M. J. Gidley. 2009. Metabolic regulation of feed intake in monogastric mammals. In: D. Torrallardona and E. Roura (eds.) Voluntary feed intake in pigs. Wageningen Academic Publishers, the Netherlands.

Boler, D. D., J. Killefer, D. M. Meeuwse, V. L. King, F. K. McKeith, and A. C. Dilger. 2012. Effects of slaughter time post-second injection on carcass cutting yields and bacon characteristics of immunologically castrated male pigs. J. Anim. Sci. 90: 334-344.

Boler, D. D., L. W. Kutzler, D. M. Meeuwse, V. L. King, D. R. Campion, F. K. McKeith, and J. Killefer. 2011. Effects of increasing lysine on carcass composition and

cutting yields of immunologically castrated male pigs. J. Anim. Sci. 89: 2189-2199.
Boler, D. D., C. L. Puls, D. L. Clark, M. Ellis, A. L. Schroeder, P. D. Matzat, J. Killefer, F. K. McKeith, and A. C. Dilger. 2014. Effects of immunological castration (Improvest) on changes in dressing percentage and carcass characteristics of finishing pigs. J. Anim. Sci. 92: 359-368.
Bonneau, M. 1982. Compounds responsible for boar taint, with special emphasis on androstenone: A review. Livest. Prod. Sci. 9: 687-705.
Bonneau, M. 2006. Factors affecting the level of androstenone. Acta Vet. Scand. 48: S7-S10.
Bonneau, M. 2010. Accessory sex glands as a tool to measure the efficacy of immunocastration in male pigs. Animal 4: 930-932.
Bonneau, M., and P. Russeil. 1985. The size of cowper's (bulbo-urethral) glands as an estimate of boar taint on the slaughter line. Livest. Prod. Sci. 13: 169-178.
Boyd, G., and R. Cady. 2012. A 50-Year Comparison fo the Carbon Footprint of the US Swine Herd: 1959 - 2009.
Boyd, R. D., M. E. Johnston, K. Scheller, A. A. Sosnicki, and E. R. Wilson. 1997. Relationship between dietary fatty acid profile and body fat composition in growing pigs, PIC, Franklin, KY.
Bradford, J. R., and M. A. Mellencamp. 2013. Immunological control of boar taint and aggressive behavior in male swine. Anim. Front. 3: 12-19.
Brooks, R. I., and A. M. Pearson. 1986. Steroid hormone pathways in the pig, with special emphasis on boar odor: A review. J. Anim. Sci. 62: 632-645.
Browne, N. A., J. K. Apple, B. E. Bass, C. V. Maxwell, J. W. S. Yancey, T. M. Johnson, and D. L. Galloway. 2013. Alternating dietary fat sources for growing-finishing pigs fed dried distillers grains with solubles I. Growth performance, pork carcass characteristics, and fatty acid composition of subcutaneous fat depots. J. Anim. Sci. 91: 1493-1508.
Brumm, M. 2012. Look for higher slaughter weights by 2022. National Hog Farmer Expert Blog.
Brunius, C., M. K. Rasmussen, H. Lacoutiére, K. Andersson, B. Ekstrand, and G. Zamaratskaia. 2011. Expression and activities of hepatic cytochrome P450 (CYP1A, CYP2A and CYP2E1) in entire and castrated male pigs. Animal 6: 271.
Bryden, W. L. 2012. Mycotoxin contamination of the feed supply chain: Implications for animal productivity and feed security. Anim. Feed Sci. Technol. 173: 134-158.
Campbell, R. G., N. C. Steele, T. J. Caperna, J. P. McMurtry, M. B. Solomon, and A. D. Mitchell. 1989. Interrelationship between sex and exogenous growth hormone administration on performance, body composition and protein and fat accretion of growing pigs. J. Anim. Sci. 67: 177-186.
Cao, Z. J., J. L. Anderson, and K. F. Kalscheur. 2009. Ruminal degradation and intestinal digestibility of dried or wet distillers grains with increasing concentrations of condensed distillers solubles. J. Anim. Sci. 87: 3013-3019.
Capper, J. L. 2011. Replacing rose-tinted spectackles with a hihg-powered microscope: the historical versus modern carbon footprint of animal agriculture. Anim. Front. 1: 26-32.

Chen, H. Y., A. J. Lewis, P. S. Miller, and J. T. Yen. 1999. The effect of excess protein on growth performance and protein metabolism of finishing barrows and gilts. J. Anim. Sci. 77: 3238-3247.

Clapper, J. A., T. M. Clark, and L. A. Rempel. 2000. Serum concentrations of IGF-1, estradiol-17beta, testosterone, and relative amnounts of IGF binding proteins (IGFBP) in growing boars, barrows, and gilts. J. Anim. Sci. 78: 2581-2588.

Claus, R., M. Lacorn, K. Danowski, M. C. Pearce, and A. Bauer. 2007. Short-term endocrine and metabolic reactions before and after second immunization against GnRH in boars. Vaccine 25: 4689-4696.

Claus, R., D. Lösel, M. Lacorn, J. Mentschel, and H. Schenkel. 2003. Effects of butyrate on apoptosis in the pig colon and its consequences for skatole formation and tissue accumulation. J. Anim. Sci. 81: 239-248.

Claus, R., S. Raab, and S. Röckle. 1996. Skatole concentrations in blood plasma of pigs as influenced by the effects of dietary factors on gut mucosa proliferation. J. Anim. Physiol. Anim. Nutr. 76: 170-179.

Claus, R., S. Rottner, and C. Rueckert. 2008. Individual return to Leydig cell function after GnRH-immunization of boars. Vaccine 26: 4571-4578.

Claus, R., and U. Weiler. 1994. Endrocrine regulation of growth and metabolism in the pig: a review. Livest. Prod. Sci. 37: 245-260.

Claus, R., U. Weiler, and A. Herzog. 1994a. Physiological aspects of androstenone and skatole formation in the boar—A review with experimental data. Meat Sci. 38: 289-305.

Claus, R., U. Weiler, and A. Herzog. 1994b. Physiological aspects of androstenone and skatole formation in the boar - a reivew with experimental data. Meat Sci. 38: 289-305.

Coble, K. F., J. M. DeRouchey, M. D. Tokach, R. D. Goodband, S. S. Dritz, T. A. Houser, B. L. Goehring, and M. J. Azain. 2013. Effects of dietary byproduct feeding withdrawal prior to market on finishing pig growth performance, carcass characteristics, carcass fat quality, intestinal weights, and economics. In: Swine Day 2013, Manhattan, KS. p 186-204.

Coker, M. D., R. L. West, J. H. Brendemuhl, D. D. Johnson, and A. M. Stelzleni. 2009. Effects of live weight and processing on the sensory traits, androstenedione concentration and 5-alpha-androst-16-en-3-one (androstenone) concentration in boar meat. Meat Sci. 82: 399-404.

Correa, J. A., C. Gariepy, M. Marcoux, and L. Faucitano. 2008. Effects of growth rate, sex and slaughter weight on fat characterisitics of pork bellies. Meat Sci. 80: 550-554.

Cromwell, G. L., M. J. Azain, O. Adeola, S. K. Baidoo, S. D. Carter, T. D. Crenshaw, S. W. Kim, D. C. Mahan, P. S. Miller, and M. C. Shannon. 2011. Corn distillers dried grains with solubles in diets for growing-finishing pigs: A cooperative study. J. Anim. Sci. 89: 2801-2811.

Cronin, G., F. Dunshea, K. Butler, I. McCauley, J. Barnett, and P. Hemsworth. 2003. The effects of immuno-and surgical-castration on the behaviour and consequently growth of group-housed, male finisher pigs. Appl. Anim. Behav. Sci. 81: 111-126.

Curry, S. M., D. Navarro, F. N. Almeida, J. A. S. Almeida, and H. H. Stein. 2014. Amino acid digestibility in low-fat distillers dried grains with solubles fed to growing pigs. J. Anim. Sci. Biotechnol. 5.

D'Souza, D. N., D. W. Pethick, F. R. Dunshea, D. Suster, J. R. Pluske, and B. P. Mullan. 2004. The pattern of fat and lean muscle tissue depostion differs in the different pork primal cuts of female pigs during the finisher growth phase. Livest. Prod. Sci. 91: 1-8.

Dahlen, R. B. A., S. K. Baidoo, G. C. Shurson, J. E. Anderson, C. R. Dahlen, and L. J. Johnston. 2011. Assessment of energy content of low-solubles corn distillers dried grains and effects on growth performance, carcass characteristics, and pork fat quality in growing-finishing pigs. J. Anim. Sci. 89: 3140-3152.

de Lange, C. F. M., S. H. Birkett, and P. C. H. Morel. 2001. Protein, Fat, and Bone Tissue Growth in Swine. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. p 65-81. CRC Press, Boca Raton.

de Lange, C. F. M., P. C. H. Morel, and S. H. Birkett. 2003. Modeling chemical and physical body composition of the growing pig. J. Anim. Sci. 81: E159-E165.

De Moraes, P. J. U., J. Allison, J. A. Robinson, G. L. Baldo, F. Boeri, and P. Borla. 2013. Life cycle assessment (LCA) and environmental production declaration (EPD) of an immunoloigcal product for boar taint control in male pigs. J. Environ. Assess. Policy Manag. 15: 1350001.

De Smet, S. 2012. Meat, poultry, and fish composition: strategies for optimizing human intake of essential nutrients. Anim. Front. 2: 10-16.

De Smet, S., K. Raes, and D. Demeyer. 2004. Meat fatty acid composition as affected by fatness and genetic factors: a review. Anim. Res. 53: 81-98.

DeRouchey, J. M., and B. T. Richert. 2010. Feeding systems for swine, U.S. Pork Center of Excellence.

Dodson, M. V., G. J. Hausman, L. L. Guan, M. Du, T. P. Rasmussen, S. P. Poulos, P. Mir, W. G. Bergen, M. E. Fernyhough, and D. C. McFarland. 2010. Lipid metabolism, adipocyte depot physiology and utilization of meat animals as experimental models for metabolic research. Int. J. Biol. Sci. 6: 691.

Doran, E., F. M. Whittington, J. D. Wood, and J. D. McGivan. 2002. Cytochrome P450IIEI (CYP2E1) is induced by skatole and this induction is blocked by androstenone in isolated pig hepatocytes. Chem. Biol. Interact. 140: 81-92.

Doran, E., F. M. Whittington, J. D. Wood, and J. D. McGivan. 2004. Characterisation of androstenone metabolism in pig liver microsomes. Chem. Biol. Interact. 147: 141-149.

Dritz, S. S., M. D. Tokach, R. D. Goodband, J. M. DeRouchey, J. L. Nelssen, D. M. Meeuwse, V. L. King, P. L. Runnels, and J. P. Crane. 2011. Effects on performance characteristics of immunologically castrated male pigs using different levels of lysine in the diets. Kansas State University and Pfizer Animal Health.

Dunshea, F., R. King, R. Campbell, R. Sainz, and Y. Kim. 1993. Interrelationships between sex and ractopamine on protein and lipid deposition in rapidly growing pigs. J. Anim. Sci. 71: 2919-2930.

Dunshea, F. R., J. R. D. Allison, M. Bertram, D. D. Boler, L. Brossard, R. Campbell, J. P. Crane, D. P. Hennessy, L. Huber, and C. de Lange. 2013. The effect of

immunization against GnRF on nutrient requirements of male pigs: A review. Animal 7: 1769-1778.
Dunshea, F. R., C. Colantoni, K. Howard, I. McCauley, P. Jackson, K. A. Long, S. Lopaticki, E. A. Nugent, J. A. Simons, J. Walker, and D. P. Hennessy. 2001. Vaccination of boars with a GnRH vaccine (Improvac) eliminates boar taint and increases growth performance. J. Anim. Sci. 79: 2524-2535.
Duttlinger, A., J. DeRouchey, M. Tokach, S. Dritz, R. Goodband, J. Nelssen, T. Houser, and R. Sulabo. 2012. Effects of increasing crude glycerol and dried distillers grains with solubles on growth performance, carcass characteristics, and carcass fat quality of finishing pigs. J. Anim. Sci. 90: 840-852.
EIA, U. S. 2014a. Petroleum and other liquids http://www.eia.gov/dnav/pet/pet_move_impcus_a2_nus_epc0_im0_mbbl_a.htm Accessed May 14, 2014.
EIA, U. S. 2014b. Petroleum and other liquids spot sprices. http://www.eia.gov/dnav/pet/pet_pri_spt_s1_d.htm Accessed October 11 2014.
Ellis, M., and N. R. Augsperger. 2001. Feed intake in growing-finishing pigs. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. p 447-467. CRC Press.
Ellis, N. R., and H. S. Isbell. 1926. Soft pork studies: II. The influence of the character of the ration upon the compositon of the body fat of hogs. J. Biol. Chem. 69: 219-238.
Elsbernd, A. J., C. F. M. De Lange, and J. F. Patience. 2014. Characterizing the feed intake pattern of immunologically castratedmale pigs following administration of the second anti-GnRF injection. J. Anim. Sci. 92 (Suppl. 2): 147 (Abstr.).
Erickson, M. C. 2008. Lipid oxidation of muscle foods. In: C. C. Akoh and D. B. Min (eds.) Food lipids: chemistry, nutrition, and biotechnology. CRC Press, Boca Raton.
Fairfield, D. 2005. Pelleting Systems and Operations. In: E. K. Schofield (ed.) Feed Manufacturing Technology. American Feed Industry Association, Arlington, VA.
Fan, M. Z. 2013. Swine nutrition and environment. In: L. I. Chiba (ed.) Sustainable Swine Nutrition. p 365-411. John Wiley & Sons, Inc., Ames.
FAO. 2013. FAO Statistical Yearbook: 2013 World Food and Agriculture. Food and Agriculture Organization of the United Nations, Rome.
Farnworth, E., and J. Kramer. 1987. Fat metabolism in growing swine: A review. Can. J. of Anim. Sci. 67: 301-318.
FAWC. 2009. Farm animal welfare in Great Britain: Past, present, future, Farm Animal Welfare Council.
FDA. 2011a. Freedom of information summary - Supplemental new animal drug application
FDA. 2011b. Freedom of Information Summary: New Animal Drug Application: Improvest.
FDA. 2011c. Freedom of Information Summary: Original New Animal Drug Application for Improvest.
Font-i-Furnols, M. 2012. Consumer studies on sensory acceptability of boar taint: A review. Meat Sci. 92: 319-329.
FSIS. 2013. FSIS Notice: Instructions for Verification of Improvest Hogs, Washington, D.C.

Gaines, A. M., J. D. Spencer, G. I. Petersen, N. R. Augsperger, and S. J. Kitt. 2007. Effect of corn distiller's dried grains with solubles (DDGS) withdrawal program on growth performance and carcass yield in grow-finish pigs. J. Anim. Sci. 88(Suppl. 1): 438(Abstr.).

Ganesan, V., K. Muthukumarappan, and K. A. Rosentrater. 2008. Effect of moisture content and soluble level on the physical, chemical, and flow properties of distillers dried drains with solubles (DDGS). Cereal Chem. 85: 464-470.

Ganesan, V., K. A. Rosentrater, and K. Muthukumarappan. 2009. Physical and flow properties of regular and reduced fat distillers dried grains with solubles (DDGS). Food Bioprocess. Technol. 2: 156-166.

Gispert, M., M. A. Oliver, A. Velarde, P. Suarez, J. Perez, and M. Font i Furnols. 2010. Carcass and meat quality characterisitics of immunocastrated male, surgically castrated male, entire male and female pigs. Meat Sci. 85: 664-670.

Godber, J. S. 1994. Nutritional vale of muscle foods. In: D. M. Kinsman, A. W. Kotula and B. C. Breidenstein (eds.) Muscle Foods. p 430-455. Chapman & Hall, Inc., New York.

Godfray, H. C. J., J. R. Beddington, I. R. Crute, L. Haddad, D. Lawrence, J. F. Muir, J. Pretty, S. Robinson, S. M. Thomas, and C. Toulmin. 2010. Food security: The challgenge of feeding 9 billion people. Science 327: 812-818.

Graham, A. B., R. D. Goodband, M. D. Tokach, S. S. Dritz, J. M. DeRouchey, S. Nitikanchana, and J. J. Updike. 2014. The effects of low-, medium-, and high-oil distillers dried grains with solubles on growth performance, nutrient digestibility, and fat quality in finishing pigs. J. Anim. Sci.: jas. 2014-7678.

Graham, H., C. Piotrowski, and R. Van Barneveld. 2013. Taking near infrared spectroscopy beyond feedstuff analysis to enhance animal production profitability. Anim. Prod. Sci. 53: 1179-1181.

Guay, K., G. Salgado, G. Thompson, B. Backus, A. Sapkota, W. Chaya, and J. J. McGlone. 2013. Behavior and handling of physically and immunoligcally castrated market pigs on farm and going to market. J. Anim. Sci. 91: 5410-5417.

Han, J., and K. Liu. 2010. Changes in composition and amino acid profile during dry grind ethanol processing from corn and estimation of yeast contribution toward DDGS proteins. J. Agric. Food Chem. 58: 3430-3437.

Hansen, L. L., A. E. Larsen, B. B. Jensen, J. Hansen-Møller, and P. Barton-Gade. 1994. Influence of stocking rate and faeces deposition in the pen at different temperatures on skatole concentration (boar taint) in subcutaneous fat. Anim. Sci. 59: 99-110.

Hardman, S. J. 2014. Effect of dietary distillers dried grains with solubles (DDGS) and pig removal strategy at harvest on the growth performance, carcass characterisitics, and fat quality of growing-finishing pigs, University of Illinois, Urbana-Champaign.

Harris, D. L. H. (Editor), 2013. Hemorrhagic Bowel Syndrome in Pigs. The Merck Veterinary Manual.

Hathaway, M. R., W. R. Dayton, M. E. White, T. L. Henderson, and T. B. Henningson. 1996. Serum insulin-like growth factor I (IGF-I) concentrations are increased in pigs fed antimicrobials. J. Anim. Sci. 74: 1541-1547.

Hilbrands, A. M., L. J. Johnston, K. M. McClelland, R. B. Cox, S. K. Baidoo, L. W. O. Souza, and G. C. Shurson. 2013. Effects of abrupt introduction and removal of high and low digestibility corn distillers dried grains with solubles from the diet on growth performance and carcass characteristics of growing-finishing pigs. J. Anim. Sci. 91: 248-258.

Hill, C., E. Raizman, T. Snider, S. Goyal, M. Torremorell, and A. M. Perez. 2014. Emergence of porcine epidemic diarrhoea in North America. In: FAO (ed.) No. FOCUS ON, No. 9. FAO, Rome.

Hill, G. M., J. E. Link, M. A. Liptrap, M. A. Giesemann, M. J. Dawes, J. A. Snedegar, N. M. Bello, and R. J. Tempelman. 2008. Withdrawal of distillers dried grains with solubles (DDGS) prior to slaughter in finishing pigs. J. Anim. Sci. 86 (e-Suppl. 3) 52(Abstr.).

Hossner, K. L. 2005. Growth hormone and insulin-like growth factors. CABI Publishing, Cambridge.

Huang, Z., I. J. Salfer, P. E. Urriola, M. D. Stern, and G. C. Shurson. 2014. Evaluation of in vitro dry matter digestibility (IVDMD) of high-fiber feed ingriedients for monogastrics. J. Anim. Sci. 92(Suppl. 2): 80-81(Abstr.).

Huber, L. 2012. Nutrient retention in entire male pigs immunized against gonadotropin-releasing hormone, The University of Guelph, Ontario, Canada.

Huff-Lonergan, E., T. J. Baas, M. Malek, J. C. Dekkers, K. Prusa, and M. F. Rothschild. 2002. Correlations among selected pork quality traits. J. Anim. Sci. 80: 617-627.

Hugo, A., and E. Roodt. 2007. Significance of porcine fat quality in meat technology: A review. Food Rev. Int. 23: 175-198.

IMPS. 2014. Institutional Meat Purchase Specifications: Fresh Pork, USDA-AMS.

INRA. 2008. EvaPig® Evaluation of pig feeds-Equations and Coefficients INRA-SENAH.

Jacela, J. Y., J. M. Benz, S. S. Dritz, M. D. Tokach, J. M. DeRouchey, R. D. Goodband, J. L. Nelssen, and K. J. Prusa. 2009. Effect of dried distillers grains with solubles withdrawal regimens on finishing pig performance and carcass characteristics Swine Day. p 181-191, Manhattan, KS.

Jacobsen, T. 2014. https://www.thejacobsen.com/about-us Accessed October 30 2014.

Jacobson, L., B. Hu, M. Yan, N. Akdeniz, and C. Clanton. 2013. The potential causes of manure pit foaming in pig finishing barns American Society of Agricultural and Biological Engineers Anual International Meeting 2013 No. 6. p 4764-4773. ASABE, Kansas City, MO.

Jarret, G., J. Martinez, and J.-Y. Dourmad. 2011. Effect of biofuel co-products in pig diets on the excretory patterns of N and C and on the subsequent ammonia and methane emissions from pig effluent. Animal 5: 622-631.

Jensen, B. B. 2006. Prevention of boar taint in pig production. Factors affecting level of skatole. Acta Vet. Scand. 48: S6-S9.

Jensen, M. T., R. P. Cox, and B. B. Jensen. 1995. Microbial production of skatole in the hind gut of pigs given different diets and its relation to skatole deposition in backfat. Anim. Sci. 61: 293-304.

Johnson, A. K., L. M. Gesing, M. Ellis, J. J. McGlone, E. Berg, S. M. Lonergan, R. Fitzgerald, L. A. Karriker, A. Ramirez, and K. J. Stalder. 2013. 2011 and 2012

early careers achievement awards:Farm and pig factors affecting welfare during the marketing process. J. Anim. Sci. 91: 2481-2491.
Kellner, T. A. 2014. Impact of dietary fat intake on carcass iodine value and pork fat quality, Iowa State University, Ames.
Kerr, B. J., W. A. Dozier, and G. C. Shurson. 2013. Effects of reduced-oil corn distillers dried grains with solubles composition on digestible and metabolizable energy value and prediction in growing pigs. J. Anim. Sci. 91: 3231-3243.
Kerr, B. J., and G. C. Shurson. 2013. Strategies to improve fiber utilization in swine. J. Anim. Sci. Biotechnol. 4: 11.
Kersten, S. 2001. Mechanisms of nutritional and hormonal regulation of lipogenesis. EMBO reports 2: 282-286.
Kil, D. Y., T. E. Sauber, D. B. Jones, and H. H. Stein. 2010. Effect of the form of dietary fat and the concentration of dietary neutral detergent fiber on ileal and total tract endogenous losses and apparent and true digestibility of fat by growing pigs. J. Anim. Sci. 88: 2959-2967.
Kim, B. G., D. Y. Kil, and H. H. Stein. 2013. In growing pigs, the true ileal and total tract digestibility of acid hydrolyzed ether extract in extracted corn oil is greater than in intact sources of corn oil or soybean oil. J. Anim. Sci. 91: 755-763.
Kim, H. J., and D. B. Min. 2008. Chemistry of lipid oxidation. In: C. C. Akoh and D. B. Min (eds.) Food lipids: Chemistry, nutrition, and biotechnology. CRC Press, Boca Raton.
Koch, D. E., A. M. Pearson, W. T. Magee, J. A. Hoefer, and B. S. Schweigert. 1968. Effect of Diet on the Fatty Acid Composition of Pork Fat. J. Anim. Sci. 27: 360-365.
Kornegay, E. T. 2001. Digestion of Phosphorus and Other Nutrients: the Role of Phytases and Factors Influencing their Activity. In: M. R. Bedford and G. G. Partridge (eds.) Enzymes in Farm Animal Nutrition. p 237-271. CAB International.
Kornegay, E. T., and M. W. A. Verstegen. 2001. Swine nutrition and environmental pollution and odor control. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. CRC Press, Boca Raton.
Kouba, M., and J. Mourot. 1998. Effect of a high linoleic acid diet on delta 9-desaturase activity, lipogenesis and lipid composition of pig subcutaneous adipose tissue. Reprod. Nutr. Dev. 38: 31-37.
Kyle, J. M., B. M. Bohrer, A. L. Schroeder, R. J. Matulis, and D. D. Boler. 2014. Effects of immunological castration (Improvest) on further processed belly characteristics and commercial bacon slicing yields of finishing pigs. J. Anim. Sci. 92: 4223-4233.
Kyriakidis, N., and T. Katsiloulis. 2000. Calculation of iodine value from measurements of fatty acid methyl esters of some oils: Comparison with the relevant American Oil Chemists Society method. J. Amer. Oil Chem. Soc. 77: 1235-1238.
Lamberson, W. R., T. J. Safranski, R. O. Bates, D. H. Keisler, and R. L. Matteri. 1995. Relationships of serum insulin-like growth factor I concentrations to growth, composition, and reproductive traits of swine. J. Anim. Sci. 73: 3241-3245.
Lawrence, T. L. J., V. R. Fowler, and J. E. Novakofski. 2012. Growth of Farm Animals. 3rd ed. CABI, Cambridge, MA.

Le Bellego, L., J. van Milgen, S. Dubois, and J. Noblet. 2001. Energy utilization of low-protein diets in growing pigs. J. Anim. Sci. 79: 1259-1271.
Lealiifano, A. K., J. R. Pluske, R. R. Nicholls, F. R. Dunshea, R. G. Campbell, D. P. Hennessy, D. W. Miller, C. F. Hansen, and B. P. Mullan. 2011. Reducing the length of time between harvest and the secondary gonadotropin-releasing factor immunization improves growth performance and clears boar taint compounds in male finishing pigs. J. Anim. Sci. 89: 2782-2792.
Lee, J. W., D. Y. Kil, B. D. Keever, J. Killefer, F. K. McKeith, R. C. Sulabo, and H. H. Stein. 2013. Carcass fat quality of pigs is not improved by adding corn germ, beef tallow, palm kernel oil, or glycerol to finishing diets containing distillers dried grains with solubles. J. Anim. Sci. 91: 2426-2437.
Leick, C. M., C. L. Puls, M. Ellis, J. Killefer, T. R. Carr, S. M. Scramlin, M. B. England, A. M. Gaines, B. F. Wolter, S. N. Carr, and F. K. McKeith. 2010. Effect of distillers dried grains with solubles and ractopamine (Paylean) on quality and shelf-life of fresh pork and bacon. J. Anim. Sci. 88: 2751-2766.
Lewis, A. J. 2001. Amino acids in swine nutrition. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. CRC Press, Boca Raton.
Liu, K. 2011. Chemical composition of distillers grains, a review. J. Agric. Food Chem. 59: 1508-1526.
Liu, K. 2012. Grain structure and composition. In: K. Liu and K. A. Rosentrater (eds.) Distillers Grains: Production, Properties, and Utilization. CRC Press, Boca Raton.
Liu, P., L. W. O. Souza, S. K. Baidoo, and G. C. Shurson. 2012. Impact of distillers dried grains with solubles particle size on nutrient digestibility, DE and ME content, and flowability in diets for growing pigs. J. Anim. Sci. 90: 4925-4932.
Lundstrom, K., K. R. Matthews, and J. E. Haugen. 2009. Pig meat quality from entire males. Animal 3: 1497-1507.
Lunstra, D., J. Ford, R. Christenson, and R. Allrich. 1986. Changes in Leydig cell ultrastructure and function during pubertal development in the boar. Biol. Reprod. 34: 145-158.
Mackay, J., M. C. Pearce, S. Thevasagayam, and O. Doran. 2013. Fatty acid composition and lipogenic enzyme protein expression in subcutaneous adipose tissue of male pigs vaccinated against boar taint, barrows, and entire boars. J. Anim. Sci. 91: 395-404.
Madsen, A., K. Jakobsen, and H. P. Mortensen. 1992. Influence of Dietary Fat on Carcass Fat Quality in Pigs. A Review. Acta Agric. Scand. Sect. A Anim. Sci. 42: 220-225.
McClelland, K. M., G. Rentfrow, G. L. Cromwell, M. D. Lindemann, and M. J. Azain. 2012. Effects of corn distillers dried grains with solubles on quality traits of pork. J. Anim. Sci. 90: 4148-4156.
McGlone, J. J., R. I. Nicholson, J. M. Hellman, and D. N. Herzog. 1993. The development of pain in young pigs associated with castration and attempts to prevent castration-induced behavioral changes. J. Anim. Sci. 71: 1441-1446.
Meadus, W., P. Duff, B. Uttaro, J. L. Aalhus, D. C. Rolland, L. L. Gibson, and M. E. R. Dugan. 2010. Production of docosahexaenoic acid (DHA) enriched bacon. J. Agric. Food Chem. 58: 465-472.

Meisinger, D. J. 2003. The national pork quality benchmarking study. In: Reciprocal Meats Conference, Columbia, MO. p 71-72.
Meyer, S., and L. Steiner. 2014a. Daily Livestock Report. http://www.dailylivestockreport.com/documents/DLR%2002-18-2014.pdf Accessed February 18, 2014 2014.
Meyer, S., and L. Steiner. 2014b. Daily Livestock Report. http://www.dailylivestockreport.com/documents/DLR%2003-12-2014.pdf Accessed August 12, 2014 2014.
Meyer, S., and L. Steiner. 2014c. Daily Livestock Report. Accessed September 3, 2014 2014.
Miller, M. F., S. D. Shackelford, K. D. Hayden, and J. O. Reagan. 1990. Determination of the alteration in fatty acid profiles, sensory characteristics and carcass traits of swine fed elevated levels of monounsaturated fats in the diet. J. Anim. Sci. 68: 1624-1631.
Millet, S., K. Gielkens, D. De Brabander, and G. P. J. Janssens. 2011. Considerations on the performance of immunocastrated male pigs. Animal 5: 1119-1123.
Moreno, R., P. Miller, T. E. Burkey, S. J. Jones, S. Cuppett, T. Carr, T. Jones, and R. Diedrichsen. 2010. The effect of corn distillers dried grains with solubles (DDGS) on carcass quality, color stability, and sensory characteristics of pork, Univeristy of Nebraska - Lincoln, Lincoln, NE.
Morgan, J. B., G. C. Smith, J. Cannon, F. K. McKeith, and J. Heavner. 1994. Pork distribution channel audit report, National Pork Producers Council, Des Moines, IA.
Morgan, J. B., G. C. Smith, S. K. Fitzgerald, J. A. Sherbeck, C. C. Kukay, W. L. Rodakovich, and S. W. Neel. 1995. International pork quality audit: A foreign market audit for U.S. pork Colorado State University, Fort Collins.
Mörlein, D., and E. Tholen. 2015. Fatty acid composition of subcutaneous adipose tissue from entire male pigs with extremely divergent levels of boar taint compounds - an exploratory study. Meat Sci. 99: 1-7.
Mourot, J., M. Kouba, and P. Peiniau. 1995. Comparative study of *in vitro* lipogenesis in various adipose tissues in the growing domestic pig (*Sus domesticus*). Comp. Biochem. Physiol. B Biochem. Mol. Biol. 111: 379-384.
Naidu, K., V. Singh, D. B. Johnston, K. D. Rausch, and M. E. Tumbleson. 2007. Effects of Ground Corn Particle Size on Ethanol Yield and Thin Stillage Soluble Solids. Cereal Chem. 84: 6-9.
NCGA. 2014. The World of Corn, National Corn Growers Association.
Nicolau-Solano, S. I., F. M. Whittington, J. D. Wood, and O. Doran. 2007. Relationship between carcass weight, adipose tissue androstenone level and expression of the hepatic 3β-hydroxysteroid dehydrogenase in entire commercial pigs. Animal 1: 1053-1059.
Noblet, J., and J. van Milgen. 2013. Energy and Engy Metabolism in Swine. In: L. I. Chiba (ed.) Sustainable Swine Nutrition. p 23-57. John Wily & Sons, Inc.
Nold, R. A., J. R. Romans, W. J. Costello, J. A. Henson, and G. W. Libal. 1997. Sensory characteristics and carcass traits of boars, barrows, and gilts fed high- or adequate-protien diets and slaughtered at 100 or 110 kilograms. J. Anim. Sci. 75: 2641-2651.

Nold, R. A., J. R. Romans, W. J. Costello, and G. W. Libat. 1999. Charactization of muscles from boars, barrows, and gilts slaughtered at 100 or 110 kilograms: differences in fat, mositure, color, water-holding capacity, and collagen. J. Anim. Sci. 77: 1746-1754.
NPB. 2013. Quick Facts: The Pork Industry at a Glance, National Pork Board, Des Moines, IA.
NPPC. 2000. Pork composition and quality assessment procedures. Natl. Pork Board, Des Moines, IA.
NRC. 2005. Phosphorus Mineral Tolerance of Animals. p 290-305. National Academies Press, Washington, D.C.
NRC. 2012. Nutrient Requirements of Swine. 11th ed. NRC Press.
NSNG. 2010. Diet Formulation Evaulation U.S. Pork Center of Excellence, Des Moines, IA.
O'Keefe, S. F. 2008. Nomenclature and classification of lipids. In: C. C. Akoh and D. B. Min (eds.) Food lipids: Chemistry, nutrition, and biotechnology. CRC Press, Boca Raton.
Pahm, A. A., C. Pedersen, D. Hoehler, and H. H. Stein. 2008. Factors affecting the variability in ileal amino acid digestibility in corn distillers dried grains with solubles fed to growing pigs. J. Anim. Sci. 86: 2180-2189.
Patience, J. F. 2012. The influence of dietary energy on feed efficiency in grow-finish swine. In: J. F. Patience (ed.) Feed Efficiency in Swine. p 101-129. Wageningen Academic Press, the Netherlands.
Paulk, C. B., J. R. Bergstrom, M. D. Tokach, S. S. Dritz, D. D. Burnett, J. M. DeRouchey, R. D. Goodband, J. L. Nelssen, and J. M. Gonzalez. 2014. Utilizing meta-analysis to generate prediction equations for pork carcass back, belly, and jowl fat iodine value. J. Anim. Sci. 92: 374.
Pauly, C., P. Spring, D. Gahan, and J. V. O'Doherty. 2011. The effect of cereal type and enzyme supplementaiton on carcass characterisitics, volatile fatty acids and intestinal microflora and boar taint in entire male pigs. Animal 5: 378-386.
Pauly, C., P. Spring, J. V. O'Doherty, S. A. Kragten, and G. Bee. 2009. Growth performance, carcass characteristics, and meat quality of group-penned surgically castrated, immunocastrated (Improvac) and entire male pigs and individually penned entire male pigs. Animal 3: 1057-1066.
Pedersen, C., M. G. Boersma, and H. H. Stein. 2007. Digestibility of energy and phosphorus in ten samples of distillers dried grains with solubles fed to growing pigs. J. Anim. Sci. 85: 1138-1176.
Person, R. C., D. R. McKenna, D. B. Griffin, F. K. McKeith, J. A. Scanga, K. E. Belk, G. C. Smith, and J. W. Savell. 2005. Benchmarking value in the pork supply chain: Processing characteristics and consumer evaluations of pork bellies of different thicknesses when manufactured into bacon. Meat Sci. 70: 121-131.
Plain, R. 2013. Economic Impact of U.S. Pork Trade, 1986-2012, University of Missouri, Columbia, MO.
Pomerenke, J., G. C. Shurson, S. K. Baidoo, and L. J. Johnston. 2011. Effects of dietary tallow and DDGS on pork fat quality. J. Anim. Sci. 89(E-Suppl. 2): 38(Abstr.).
Prusa, K., H. Nederveld, P. L. Runnels, R. Li, V. L. King, and J. P. Crane. 2011. Prevalence and relationships of sensory taint, 5α-androstenone and skatole in fat

and lean tissue from the loin (Longissimus dorsi) of barrows, gilts, sows, and boars from selected abattoirs in the United States. Meat Sci. 88: 96-101.
Puls, C. L., A. Rojo, M. Ellis, D. D. Boler, F. K. McKeith, J. Killefer, A. M. Gaines, P. D. Matzat, and A. L. Schroeder. 2014. Growth performance of immunologically-castrated (with Improvest) barrows (with or without ractopamine) compared to gilt, physically-castrated barrow, and intact male pigs. J. Anim. Sci. 92: 2289-2295.
Raab, S., R. Leiser, H. Kemmer, and R. Claus. 1998. Effects of energy and purines in the diet on proliferation, differentiation, and apoptosis in the small intestine of the pig. Metab. 47: 1105-1111.
RFA. 2014a. Industry Resources: Co-products. http://www.ethanolrfa.org/pages/industry-resources-coproducts Accessed May 4, 2014.
RFA. 2014b. Pocket guide to ethanol 2014. In: R. F. Association (ed.). RFA.
RFA. 2014c. Renewable Fuel Standard. http://www.ethanolrfa.org/pages/renewable-fuel-standard/ Accessed April 29, 2014.
Rojas, O. J., Y. Liu, and H. H. Stein. 2013. Phosphorus digestibility and concentration of digestible and metabolizable energy in corn, corn coproducts, and bakery meal fed to growing pigs. J. Anim. Sci. 91: 5326-5335.
Rosentrater, K. A. 2003. Performance of an electrostatic dust collection system in swine facilities. Agric. Eng. Int.: CIGR J. Sci. Res. Dev. Manuscript BC 03 003.
Rosentrater, K. A. 2012. Physical properties of DDGS. In: K. Liu and K. A. Rosentrater (eds.) Distillers grains: Production, properties, and utilization. CRC Press, Boca Raton.
Rosentrater, K. A., K. Ileleji, and D. B. Johnston. 2012. Manufacturing of Fuel Ethanol and Distillers Grains - Current and Evolving Processes. In: K. Liu and K. A. Rosentrater (eds.) Distillers Grains Prodution, Properties, and Utilization. p 73-102. CRC Press, Boca Raton.
Rosenvold, K., B. Essen-Gustavsson, and H. J. Andersen. 2003. Dietary manipulation of pro- and marcoglycogen in porcine skeletal muscle. J. Anim. Sci. 81: 130-134.
Rosenvold, K., J. S. Petersen, H. N. Lwerke, S. K. Jensen, M. Therkildsen, A. H. Karlsson, H. S. Møller, and H. J. Andersen. 2001. Muscle glycogen stores and meat quality as affected by strategic finishing feeding of slaughter pigs. J. Anim. Sci. 79: 382-391.
Salfen, B. E., J. A. Carroll, and D. H. Keisler. 2003. Endocrine responses to short-term feed deprivation in weanling pigs. Journal of Endocrinology 178: 541-551.
Sather, A. P., S. D. M. Jones, W. M. Robertson, and S. Zawadski. 1995. Sex effects on fat hardness meter readings of market weight pigs. Can. J. of Anim. Sci. 75: 509-515.
Schulz, L. 2014. Estimated Livestock Returns. Estimated Livestock Returns. Iowa State University Department of Economics, Ames, IA.
Senger, P. L. 2003. Pathways to Pregnancy and Parturition. 2nd ed. Current Conceptions, Inc., Pullman, WA.
Shahidi, F., and P. K. J. P. D. Wanasundara. 2008a. Extraction and analysis of lipids. In: C. C. Akoh and D. B. Min (eds.) Food lipids: Chemistry, nutrition, and biotechnology. CRC Press, Boca Raton.

Shahidi, F., and U. N. Wanasundara. 2008b. Methods for measure oxidative rancidity in fats and oils. In: C. C. Akoh and D. B. Min (eds.) Food lipids: Chemistry, nutrition, and biotechnology. CRC Press, Boca Raton.

Shoen, T. 2014. Distillers corn oil: An alternative energy source for swine producers Feed Management.

Sinclair, P. A., E. J. Squires, and J. I. Raeside. 2001a. Early postnatal plasma concentrations of testicular steroid hormones, pubertal development, and carcass leanness as potential indicators of boar taint in market weight intact male pigs. J. Anim. Sci. 79: 1868-1876.

Sinclair, P. A., E. J. Squires, J. I. Raeside, J. H. Britt, and V. G. Hedgpeth. 2001b. The effect of early postnatal treatment with a gonadotropin-releasing hormone agonist on the developmental profiles of testicular steroid hormones in the intact male pig. J. Anim. Sci. 79: 1003-1010.

Song, R. 2013. Lipid peroxidation in corn dried distillers grains with solubles (DDGS) and effects of feeding a highly oxidized DDGS source to swine, University of Minnesota, St. Paul.

Sørensen, K., H. Petersen, and S. B. Engelsen. 2012. An on-line near-infrared (NIR) transmission method for determining depth profiles of fatty acid composition and iodine value in porcine adipose fat tissue. Applied spectroscopy 66: 218-226.

Spiehs, M. J., M. H. Whitney, and G. C. Shurson. 2002. Nutrient database for distiller's dried grains with solubles produced from new ethanol plants in Minnesota and South Dakota. J. Anim. Sci. 80: 2639-2645.

Squire, J. M., C. L. Zhu, E. A. Jeaurond, and C. F. M. de Lange. 2005. Condensed corn distillers' solubles in swine liquid feeding: growth performance and carcass quality. J. Anim. Sci. 83 (Suppl. 1): 165 (Abstr.).

Squires, E. J. 2011. Applied Animal Endocrinology. 2nd ed. CABI.

Stalder, K. J. 2012. Pork Industry Productivey Analysis, National Pork Board, Des Moines.

Stein, H. H. 2012. Feeding Ethanol Coproducts to Swine. In: K. Liu and K. A. Rosentrater (eds.) Distillers Grains: Production, Properties, and Utilization. p 297-315. CRC Press, Boca Raton.

Stein, H. H., M. F. Fuller, P. J. Moughan, B. Seve, R. Mosenthin, A. J. M. Jansman, J. A. Fernandez, and C. F. M. de Lange. 2007. Definition of apparent, true, and standardized ileal digestibility of amino acids in pigs. Livestock Science 109: 282-285.

Stein, H. H., and G. C. Shurson. 2009. The use and application of distillers dried grains with solubles in swine diets. J. Anim. Sci. 87: 1292-1303.

Suster, D., B. J. Leury, D. J. Kerton, M. R. Borg, K. L. Butler, and F. R. Dunshea. 2006. Longitudinal DXA measurements demonstrate lifetime differences in lean and fat tissue deposition between boars and barrows under individual and grou-penned systems. Aust. J. Agric. Res. 57: 1009-1015.

Sutherland, M. A., B. L. Davis, T. A. Brooks, and J. J. McGlone. 2010. Physiology and behavior of pigs before and after castration: Effects of two topical anesthetics. Animal 4: 2071-2079.

Swanson, D., R. Block, and S. A. Mousa. 2012. Omega-3 Fatty Acids EPA and DHA: Health Benefits Throughout Life. Adv. Nutr. 3: 1-7.

Tavárez, M., B. Bohrer, M. Asmus, A. Schroeder, R. Matulis, D. Boler, and A. Dilger. 2014a. Effects of immunological castration and distiller's dried grains with solubles on carcass cutability and commercial bacon slicing yields of barrows slaughtered at two time points. J. Anim. Sci. 92: 3149-3160.

Tavárez, M., C. Puls, A. Schroeder, and A. Dilger. 2014b. Effects of immunocastration and time after second Improvest dose on adipose tissue fatty acid profile of finishing barrows. J. Anim. Sci. 92: 3736-3744.

Testroet, E. D., C. Yoder, C. Bustos, S. M. Lei, D. C. Beitz, and T. J. Baas. 2014. Relationship of fat quality to meat quality traits of pork. J. Anim. Sci. 92: 210 (Abstr.).

Trusell, K. A., J. K. Apple, J. W. S. Yancey, T. M. Johnson, D. L. Galloway, and R. J. Stackhouse. 2011. Compositional and instrumental firmness variation within fresh pork bellies. Meat Sci. 88: 472-480.

Urriola, P. E., S. K. Cervantes-Pahm, and H. H. Stein. 2013a. Fiber in swine nutrition. In: L. I. Chiba (ed.) Sustainable Swine Nutrition. p 255-276. John Wiley & Sons, Inc., Ames.

Urriola, P. E., L. J. Johnston, H. H. Stein, and G. C. Shurson. 2013b. Prediction of the concentration of standardized ileal digestible amino acids in distillers dried grains with solubles. J. Anim. Sci. 91: 4389-4396.

Urriola, P. E., M. Li, B. J. Kerr, and G. C. Shurson. 2014. Evaluation of prediction equations to estimate gross, digestible, and metabolizable energy content of maize dried distillers grains with solubles (DDGS) for swine based on chemical composition. Anim. Feed Sci. Technol. In Press.

USDA-ERS. 2010. Market issues and prospects for U.S. Distillers' Grains: Supply, use, and price relationships, USDA.

USDA-ERS. 2012. Hog & Pork Trade. http://www.ers.usda.gov/topics/animal-products/hogs-pork/trade.aspx#.U20lBFU7u-0 Accessed May 9 2014.

USDA. 2014a. Crop Production 2013 Summary, USDA, Washington, D.C.

USDA. 2014b. Livestock slaughter 2013 summary USDA.

USDA. 2014c. USDA Agricultural Projections to 2023. No. (OCE-141), USDA, Washington, D.C.

USGC. 2012. A guide to distiller's dried grains with solubles (DDGS).

USGC. 2014. Buying and Selling DDGS. http://www.grains.org/buyingselling/ddgs Accessed 10/6/2014.

USMEF. 2013. Beef, pork exports set new records in 2012.

USMEF. 2014. Leading Markets for U.S. Pork Variety Meat Exports.

van Milgen, J., and J. Noblet. 2003. Partitioning of energy intake to heat, protein, and fat in growing pigs. J. Anim. Sci. 81: E86-E93.

van Milgen, J., J. Noblet, and S. Dubois. 2001. Energetic efficiency of starch, protein and lipid utilization in growing pigs. J. Nutr. 131: 1309-1318.

van Milgen, J., A. Valancogne, S. Dubois, J. Dourmad, B. Seve, and J. Noblet. 2008. InraPorc: A model and decision support tool for the nutrition of growing pigs. Anim. Feed Sci. Technol. 143: 387-405.

Wade, G. N., and J. M. Gray. 1979. Gonadal effects on food intake and adiposity: A metabolic hypothesis. Physiol. Behav. 22: 583-593.

Wade, G. N., and I. Zucker. 1970. Modulation of food intake and locomotor activity in female rats by diencephalic hormone implants. J. Comp. Physiol. Psychol. 72: 328-336.

Walstra, P., C. Claudi-Magnussen, P. Chevillon, G. von Seth, A. Diestre, K. R. Matthews, D. B. Homer, and M. Bonneau. 1999. An international study on the importance of androstenone and skatole for boar taint: levels of androstenone and skatole by country and season. Livest. Prod. Sci. 62: 15-28.

Warnants, N., M. J. Van Oeckel, and C. V. Boucqué. 1999. Incorporation of dietary polyunsaturated fatty acids into pork fatty tissues. J. Anim. Sci. 77: 2478-2490.

Weber, E., J. F. Patience, and K. J. Stalder. 2013. Feeder space availability and dried distillers grains with solubles inclusion level interaction on grow-finish pig peformance and total tract digestibility in a commercial setting, Iowa State University, Ames, IA.

Weldon, W. 2013. Enriching people's lives: A 2013 report on the importance of animal source foods. In: Midwest Swine Nutrition Conference Proceedings, Indianapolis. p 3-9.

Wesoly, R., and U. Weiler. 2012. Nutritional influences on skatole formation and skatole metabolism in the pig. Animals 2: 221-242.

White, H. M., B. T. Richert, and M. A. Latour. 2013. Impacts of Nutrition and Environmental Stressors on Lipid Metabolism. In: R. V. Baez (ed.) Lipid Metabolism. InTech.

Whitney, M. H., G. C. Shurson, L. J. Johnston, D. M. Wulf, and B. C. Shanks. 2006. Growth performance and carcass characteristics of grower-finisher pigs fed high-quality corn distillers dried grain with solubles originiating from a modern Midwestern ethanol plant. J. Anim. Sci. 84: 3356-3363.

Whittington, F. M., D. Zammerini, G. R. Nute, A. Baker, S. I. Hughes, and J. D. Wood. 2011. Comparison of heating methods and the use of different tissues for sensory assessment of abnormal odours (boar taint) in pig meat. Meat Sci. 88: 249-255.

Widmer, M., L. McGinnis, D. Wulf, and H. Stein. 2008. Effects of feeding distillers dried grains with solubles, high-protein distillers dried grains, and corn germ to growing-finishing pigs on pig performance, carcass quality, and the palatability of pork. J. Anim. Sci. 86: 1819-1831.

Wiegand, B. R., R. B. Hinson, M. J. Ritter, S. N. Carr, and G. L. Allee. 2011. Fatty acid profiles and iodine value correlations between four carcass fat depots from pigs fed varied combinations of ractopamine (Paylean) and energy. J. Anim. Sci. 89: 3580-3586.

Winkler-Moser, J. K. 2012. Lipids in DDGS. In: K. Liu and K. A. Rosentrater (eds.) Distillers Grains: Production, Properties, and Utilization. CRC Press, Boca Raton, FL.

Wise, T., D. D. Lunstra, G. A. Rohrer, and J. J. Ford. 2003. Relationships of testicular iron and ferritin concentrations with testicular weight and sperm production in boars. J. Anim. Sci. 81: 503-511.

Wood, J., R. Richardson, G. Nute, A. Fisher, M. Campo, E. Kasapidou, P. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: a review. Meat Sci. 66: 21-32.

Wood, J. D., M. Enser, A. V. Fisher, G. R. Nute, P. R. Sheard, R. I. Richardson, S. I. Hughes, and F. M. Whittington. 2008. Fat deposition, fatty acid composition and meat quality: A review. Meat Sci. 78: 343-358.

Wood, J. D., M. Enser, F. M. Whittington, C. B. Moncrieff, and A. J. Kempster. 1989. Backfat composition in pigs: Differences between fat thickness groups and sexes. Livest. Prod. Sci. 22: 351-362.

Wood, J. D., P. D. Warriss, and M. B. Enser. 1992. Effects of production factors on meat quality in pigs. In: D. A. Ledward, D. E. Johnston and M. K. Knight (eds.) The chemistry of muscle-based foods Redwood Press Ltd., Wiltshire.

Woyengo, T. A., E. Beltranena, and R. T. Zijlstra. 2014. Nonruminant nutrition symposium: Controlling feed cost by including alternative ingredients into pig diets: A review. J. Anim. Sci. 92: 1293-1305.

Wray-Cahen, D. 2001. Performance-Enhancing Substances. In: A. J. Lewis and L. L. Southern (eds.) Swine Nutrition. p 427-446. CRC Press, Boca Raton.

Wu, F., L. J. Johnston, P. E. Urriola, A. M. Hilbrands, and G. C. Shurson. 2014. Validation of ME prediction equations and the impact of feeding diets containing corn distillers dried grains with solubles (DDGS) with variable oil content on growth performance of growing-finishing pigs J. Anim. Sci. 92(Suppl. 2): 30(Abstr.).

Xu, G., S. K. Badioo, L. J. Johnston, D. Bibus, J. E. Cannon, and G. C. Shurson. 2010a. The effects of feeding diets containing corn distillers dried grains with solubles, and withdrawal period of distillers dried grains with solubles, on growth performance and pork quality in grower-finisher pigs. J. Anim. Sci. 88: 1388-1397.

Xu, G., S. K. Baidoo, L. J. Johnson, D. Bibus, J. E. Cannon, and G. C. Shurson. 2010b. Effects of feeding diets containing increasing content of corn distillers dried grains with solubles to grower-finisher pigs on growth performance, carcass composition, and pork fat quality. J. Anim. Sci. 88: 1398-1410.

Xu, G., G. He, S. K. Baidoo, and G. C. Shurson. 2006. Effect of feeding diets containing corn distillers dried grains with solubles (DDGS), with or without phytase, on nutrient digestibility and excretion in nursery pigs. J. Anim. Sci. 85(Suppl. 2): 76. (Abstr.).

Xue, J., G. D. Dial, E. E. Holton, Z. Vickers, E. J. Squires, Y. Lou, D. Godbout, and N. Morel. 1996. Breed differences in boar taint: Relationship between tissue levels boar taint compounds and sensory analysis of taint. J. Anim. Sci. 74: 2170-2177.

Yan, M. 2014. Dangerous swine manure foaming and its relations to corn ethanol co-products. In: 36th Symposium on Biotechnology for Fuels and Chemicals (April 28-May 1, 2014)

Yao, Y., Z. Cai, L. Zhang, C. Zhao, K. Wu, N. Xu, G. Liu, and C. Wu. 2009. Effects of castration on androgen receptor, IGF-1 Ea, MGF and myostatin gene expression in skeletal muscles of male pigs. Asian Austral. J. Anim. Sci. 8: 1069-1077.

Yao, Y. C., Z. W. Cai, C. J. Zhao, K. L. Wu, C. X. Wu, W. P. Han, and N. Y. Xu. 2011. Influence of castration-induced sex hormone deficiency on serum lipid levels and the genes expression in male pigs. Horm. Metab. Res. 43: 674-680.

Yuan, Y. L., J. L. Li, W. H. Zhang, C. Li, F. Gao, and G. H. Zhou. 2012. A comparison of slaughter performance and meat quality of pigs immunised with a

gonadotropin-releasing factor vaccine against boar taint with physically castrated pigs. Anim. Prod. Sci. 52: 911-916.
Zamaratskaia, G., H. K. Andersson, G. Chen, K. Andersson, A. Madej, and K. Lundstrom. 2008a. Effect of a gonadtropin-releasing hormone vaccine (Improvac) on steroid hormones, boar taint compounds and performance in entire male pigs. Reprod. Domest. Anim. 43: 351-359.
Zamaratskaia, G., J. Babol, A. Madej, E. J. Squires, and K. Lundstrom. 2004. Age-related variation of plasma concentrations of skatole, androstenone, testosterone, oestradiol-17β, oestrone sulphate, dehydroepiandrosterone sulphate, triiodothyronine and IGF-1 in six entire male pigs. Reprod. Domest. Anim. 39: 168-172.
Zamaratskaia, G., L. Rydhmer, H. K. Andersson, G. Chen, S. Lowagie, K. Andersson, and K. Lundstrom. 2008b. Long-term effect of vaccination against gonadotropin-releasing hormone, using Improvac, on hormonal profile and behaviour of male pigs. Anim. Reprod. Sci. 108: 37-48.
Zamaratskaia, G., and E. J. Squires. 2009. Biochemical, nutritional and genetic effects on boar taint in entire male pigs. Animal 3: 1508-1521.
Zhang, Y., and J. Caupert. 2011. Survey of mycotoxins in U.S. distiller's dried grains with solubles from 2009 to 2011. J. Agric. Food Chem. 60: 539-543.

Appendix

Table A.1. Interactive least squares means of overall growth performance of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies[1]

	Interval between second Improvest® dose and harvest (TD)[2]		
Trait	TD9	TD7	TD5
ADFI[3], kg/head/d			
PCon	2.45[x]	2.40	2.35
SD	2.40	2.35	2.28
WD	2.44[x]	2.40	2.34
NCon	2.31[y]	2.43	2.28
ADG[4], kg/head/d			
PCon	0.958	0.956	0.965
SD	0.935	0.943	0.942
WD	0.932	0.925	0.935
NCon	0.894	0.922	0.917
G:F[5], kg/kg			
PCon	0.420	0.426	0.435
SD	0.414	0.425	0.432
WD	0.408	0.410	0.424
NCon	0.410	0.407	0.422

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] $P < 0.10$; SEM = 0.08 kg/d.

[4] $P > 0.05$; SEM = 0.03 kg/d.

[5] $P > 0.05$; SEM = 0.006 kg/kg.

[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

Table A.2. Interactive least squares means for average daily feed intake and gain efficiency of immunologically castrated pigs when using corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 3 different time intervals after the second Improvest® dose

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
ADFI[3], kg/head/d			
8 to 11 wk	1.24^{A}	1.23^{A}	1.24^{A}
11 to 13 wk	1.72^{B}	1.72^{B}	1.72^{B}
13 to 15 wk	1.99^{B}	2.03^{BC}	2.03^{BC}
15 to 17 wk	2.44^{C}	2.35^{CD}	2.36^{CD}
17 to 19 wk	2.95^{aD}	2.64^{abD}	2.48^{bDE}
19 to 21 wk	3.25^{aD}	3.37^{aE}	2.84^{bE}
21 to 24 wk	3.20^{D}	3.39^{E}	3.51^{F}
G:F[4]			
8 to 11 wk	0.559^{A}	0.566^{A}	0.564^{A}
11 to 13 wk	0.486^{B}	0.480^{B}	0.491^{B}
13 to 15 wk	0.422^{C}	0.436^{CX}	0.434^{C}
15 to 17 wk	0.435^{C}	0.395^{CY}	0.417^{CD}
17 to 19 wk	0.358^{aD}	0.401^{bC}	0.388^{abD}
19 to 21 wk	0.338^{aD}	0.351^{aD}	0.397^{bCD}
21 to 24 wk	0.291^{E}	0.397^{E}	0.305^{E}

[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] FS × TD × Wk ($P \geq 0.21$); Wk ($P < 0.001$).

[3] TD × Wk ($P < 0.001$); SEM 0.10 kg/d.

[4] TD × Wk ($P < 0.001$); SEM 0.005.

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.3. Interactive least squares means for average daily feed intake and gain efficiency of immunologically castrated pigs when using corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 3 different time intervals after the second Improvest® dose

	Feeding Strategy (FS)[1,2]			
Trait	PCon	SD	WD	NCon
ADFI[3], kg/head/d				
8 to 11 wk	1.27^{A}	1.23^{A}	1.22^{A}	1.23^{A}
11 to 13 wk	1.77^{B}	1.71^{B}	1.70^{B}	1.70^{B}
13 to 15 wk	2.06^{C}	2.01^{C}	1.99^{C}	2.01^{C}
15 to 17 wk	2.46^{D}	2.36^{D}	2.35^{D}	2.36^{D}
17 to 19 wk	2.74^{E}	2.68^{E}	2.70^{E}	2.65^{E}
19 to 21 wk	3.14^{abF}	3.10^{bF}	3.26^{aEX}	3.11^{bF}
21 to 24 wk	3.35^{bF}	3.32^{bF}	3.51^{aY}	3.30^{bF}
G:F[4]				
8 to 11 wk	0.586^{axA}	0.556^{bA}	0.552^{bA}	0.558^{byA}
11 to 13 wk	0.488^{B}	0.499^{B}	0.476^{B}	0.479^{B}
13 to 15 wk	0.448^{C}	0.431^{C}	0.422^{C}	0.422^{C}
15 to 17 wk	0.417^{CD}	0.427^{C}	0.417^{C}	0.401^{CD}
17 to 19 wk	0.396^{D}	0.384^{D}	0.374^{D}	0.376^{DE}
19 to 21 wk	0.353^{E}	0.369^{D}	0.362^{D}	0.364^{E}
21 to 24 wk	0.298^{F}	0.301^{E}	0.292^{E}	0.290^{F}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] FS × TD × Wk ($P \geq 0.21$); Wk ($P < 0.001$).

[3] FS × Wk ($P < 0.001$); SEM 0.09 kg/d.

[4] FS × Wk ($P < 0.001$); SEM 0.008

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.4. Interactive least squares means of body weight and average daily gain of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Feeding Strategy (FS)[1,2]			
Trait	PCon	SD	WD	NCon
ADG[3], kg/head/d				
8 to 11 wk	0.74^{A}	0.68^{A}	0.68^{A}	0.69^{A}
11 to 13 wk	0.86^{B}	0.85^{B}	0.81^{B}	0.82^{B}
13 to 15 wk	0.92^{axBC}	0.87^{abB}	0.84^{bB}	0.84^{byB}
15 to 17 wk	1.03^{xDE}	1.02^{xyC}	0.98^{xyC}	0.95^{yC}
17 to 19 wk	1.08^{aDE}	1.02^{abC}	1.01^{abC}	1.00^{bC}
19 to 21 wk	1.09^{aEY}	1.14^{abD}	1.18^{bD}	1.12^{abD}
21 to 24 wk	0.99^{CDX}	1.00^{C}	1.02^{C}	0.96^{C}
Initial BW[4], kg				
Phase 1 – 8 wk	21.5^{A}	21.5^{A}	21.5^{A}	21.5^{A}
Phase 2 – 11 wk	36.8^{B}	35.6^{B}	35.4^{B}	35.6^{B}
13 wk	49.1^{C}	47.7^{C}	46.9^{C}	47.3^{C}
Phase 3 – 15 wk	62.2^{aD}	60.1^{abD}	59.0^{bD}	59.3^{bD}
17 wk	76.3^{aE}	74.0^{abE}	72.4^{bE}	72.3^{bE}
Phase 4 – 19 wk	91.7^{aF}	88.6^{bF}	86.8^{bF}	86.4^{bF}
21 wk	107.0^{aG}	104.5^{abG}	103.3^{bG}	102.1^{bG}
Final BW – 24 wk	125.3^{aH}	123.1^{abH}	122.3^{bcH}	120.0^{cH}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] FS × TD × Wk ($P \geq 0.24$); Wk ($P < 0.001$).

[3] FS × Wk ($P < 0.001$); SEM = 0.03 kg/d.

[4] FS × Wk ($P < 0.001$); SEM = 1.2 kg.

[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

[A,B,C,D,E,F,G,H] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.5. Interactive least squares means of body weight and average daily gain of immunologically castrated pigs harvested 5, 7, or 9 weeks after the second Improvest® dose while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
ADG[3], kg/head/d			
8 to 11 wk	0.70^{A}	0.70^{A}	0.70^{A}
11 to 13 wk	0.84^{B}	0.83^{B}	0.84^{BX}
13 to 15 wk	0.84^{B}	0.88^{B}	0.88^{BC}
15 to 17 wk	1.06^{aCY}	0.93^{bD}	1.00^{abCD}
17 to 19 wk	1.06^{CY}	1.06^{C}	0.96^{CDY}
19 to 21 wk	1.09^{C}	1.18^{E}	1.12^{E}
21 to 24 wk	0.93^{aBX}	0.98^{abCD}	1.07^{bDE}
Initial BW[4], kg			
Phase 1 – 8 wk	21.5	21.4	21.5
Phase 2 – 11 wk	35.8	35.8	35.9
13 wk	47.7	47.5	47.9
Phase 3 – 15 wk	59.7	60.2	60.5
17 wk	74.2	72.8	74.2
Phase 4 – 19 wk	89.2	88.0	87.9
21 wk	104.6	104.5	103.5
Final BW – 24 wk	121.9	122.7	123.4

[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] FS × TD × Wk ($P \geq 0.05$); Wk ($P < 0.001$).

[3] TD × Wk ($P < 0.001$); SEM = 0.04 kg/d.

[4] TD × Wk ($P > 0.05$); SEM = 1.2 kg.

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[A,B,C,D,E] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.6. Interactive least squares means of overall metabolizable energy (ME) and standardized ileal digestible (SID) CP and lysine intake and ME efficiency of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies[1]

	Interval between second Improvest® dose and harvest (TD)[2]		
Trait	TD9	TD7	TD5
ME intake[3,4], Mcal/head/d			
PCon	8.15	7.96	7.83
SD	7.86	7.72	7.49
WD	7.97	7.86	7.64
NCon	7.48	7.84	7.37
ME efficiency[5,6]			
PCon	8.36	8.19	7.98
SD	8.28	8.02	7.81
WD	8.35	8.27	7.98
NCon	8.24	8.33	7.91
SID CP[7,8], g/head/d			
PCon	363.5^{cdefz}	356.3^{def}	351.5^{ef}
SD	362.7^{cdefz}	353.5^{ef}	345.0^{fk}
WD	382.0^{abc}	375.3^{bcd}	365.5^{bcdej}
NCon	380.0^{aby}	402.1^{aw}	383.8^{bcx}
SID Lys[9.10], g/head/d			
PCon	21.44^{aw}	20.99^{abcj}	20.71^{abcd}
SD	20.94^{abcj}	20.41^{abcd}	19.89^{bcd}
WD	21.01^{aby}	20.69^{abcd}	20.15^{bcdx}
NCon	19.89^{cdz}	20.82^{abcd}	19.70^{dzk}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] $P < 0.10$; SEM = 0.27 Mcal/head/d.

[4] ME intake calculated as (total feed intake × calculated dietary ME density)/(pigs per pen × d on feed).

[5] $P > 0.05$; SEM = 0.11.

[6] For each period, ME efficiency was calculated as (total dietary ME intake/ total BW gain).

[7] $P < 0.05$; SEM = 11.9 g/head/d.

[8] SID CP intake calculated as (total feed intake × calculated dietary SID CP content)/(pigs per pen × d on feed).
[9] $P < 0.10$; SEM = 0.66 g/head/d.
[10] SID Lys intake calculated as (total feed intake × calculated dietary SID Lys content)/(pigs per pen × d on feed).
[a,b,c,d,e,f] Means without a common superscript differ ($P \leq 0.05$).
[w,x] Means without a common superscript differ ($P \leq 0.10$).
[y,z] Means without a common superscript differ ($P \leq 0.10$).
[j,k] Means without a common superscript differ ($P \leq 0.10$).

Table A.7. Interactive least squares means of metabolizable energy intake and efficiency of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

	Feeding Strategy (FS)[1,2]			
Trait	PCon	SD	WD	NCon
ME Intake[3,4] Mcal/head/d				
8 to 11 wk	4.22^{A}	3.96^{A}	3.94^{A}	3.96^{A}
11 to 13 wk	5.88^{B}	5.55^{B}	5.49^{B}	5.50^{B}
13 to 15 wk	6.85^{xC}	6.55^{C}	6.43^{yC}	6.48^{C}
15 to 17 wk	8.19^{axD}	7.76^{byD}	7.60^{bD}	7.63^{bD}
17 to 19 wk	9.10^{aE}	8.78^{abE}	8.74^{abE}	8.57^{bE}
19 to 21 wk	10.45^{axF}	10.27^{aF}	10.88^{byFX}	10.08^{aF}
21 to 24 wk	11.15^{aF}	10.98^{abF}	11.70^{cFY}	10.69^{bF}
ME efficiency[5,6]				
8 to 11 wk	5.67	5.80	5.84	5.77
11 to 13 wk	6.84	6.54	6.80	6.78
13 to 15 wk	7.46	7.58	7.69	7.70
15 to 17 wk	8.02	7.67	7.80	8.07
17 to 19 wk	8.48	8.63	8.67	8.63
19 to 21 wk	9.50	9.06	9.25	9.01
21 to 24 wk	11.25	10.98	11.36	11.15

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] FS × TD × Wk ($P > 0.21$); Wk ($P < 0.001$).

[3] FS × Wk ($P < 0.001$); SEM 0.29 Mcal/head/d.

[4] ME intake calculated as (total feed intake × calculated dietary ME density)/(pigs per pen × d on feed).

[5] FS × Wk ($P > 0.05$); SEM 0.17.

[6] For each period, ME efficiency calculated as (total dietary ME intake/ total BW gain).

[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.8. Interactive least squares means of metabolizable energy intake and efficiency of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
ME Intake[3,4], Mcal/head/d			
8 to 11 wk	4.03^{A}	4.00^{A}	4.02^{A}
11 to 13 wk	5.61^{B}	5.61^{B}	5.59^{B}
13 to 15 wk	6.50^{B}	6.62^{BC}	6.61^{BC}
15 to 17 wk	7.97^{C}	7.70^{CD}	7.73^{CD}
17 to 19 wk	9.64^{aD}	8.64^{abD}	8.12^{bDW}
19 to 21 wk	10.72^{aD}	11.14^{aE}	9.40^{bEX}
21 to 24 wk	10.58^{D}	11.20^{E}	11.61^{F}
ME efficiency[5,6]			
8 to 11 wk	5.81^{X}	5.74^{A}	5.76^{W}
11 to 13 wk	6.73^{AY}	6.82^{B}	6.67^{XY}
13 to 15 wk	7.75^{B}	7.52^{BC}	7.55^{AZ}
15 to 17 wk	7.55^{AB}	8.33^{C}	7.79^{AB}
17 to 19 wk	9.16^{aC}	8.19^{bC}	8.46^{abBJ}
19 to 21 wk	9.80^{aC}	9.46^{aD}	8.36^{bBH}
21 to 24 wk	11.34^{D}	11.38^{E}	10.83^{C}

[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] FS × TD × Wk ($P > 0.21$); Wk ($P < 0.001$).

[3] TD × Wk ($P < 0.001$); SEM 0.34 Mcal/head/d

[4] ME intake calculated as (total feed intake × calculated dietary ME density)/(pigs per pen × d on feed).

[5] TD × Wk ($P <0.001$); SEM 0.19

[6] For each period, ME efficiency calculated as (total dietary ME intake/ total BW gain).

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).

[W,X] Within a column, means without a common superscript differ ($P \leq 0.10$).

[Y,Z] Within a column, means without a common superscript differ ($P \leq 0.10$).

[J,H] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.9. Interactive least squares means of standardized ileal digestible (SID) CP and Lys intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Feeding Strategy (FS)[1,2]			
Trait	PCon	SD	WD	NCon
SID CP Intake[3,4], g/head/d				
8 to 11 wk	244.7^{A}	232.6^{A}	231.7^{A}	233.0^{A}
11 to 13 wk	291.3^{B}	284.5^{B}	294.4^{B}	295.0^{B}
13 to 15 wk	339.4^{C}	335.7^{C}	344.5^{C}	347.4^{C}
15 to 17 wk	353.6^{aC}	352.4^{aC}	385.4^{bD}	387.3^{bD}
17 to 19 wk	393.1^{aD}	398.8^{aD}	443.3^{bE}	435.0^{bE}
19 to 21 wk	422.7^{aE}	419.4^{aD}	443.5^{aE}	494.6^{bF}
21 to 24 wk	454.7^{aF}	452.6^{aE}	477.0^{aF}	528.3^{bG}
SID Lys Intake[5,6], g/head/d				
8 to 11 wk	13.97^{A}	13.17^{A}	13.12^{A}	13.19^{A}
11 to 13 wk	16.97^{B}	16.06^{B}	15.83^{B}	15.87^{B}
13 to 15 wk	19.77^{C}	18.95^{C}	18.53^{C}	18.68^{C}
15 to 17 wk	22.31^{D}	21.20^{D}	20.82^{D}	20.92^{D}
17 to 19 wk	23.27^{DE}	22.50^{D}	22.46^{E}	22.04^{D}
19 to 21 wk	24.59^{E}	24.54^{E}	25.80^{F}	24.31^{E}
21 to 24 wk	26.45^{abF}	26.48^{abF}	27.75^{aG}	25.97^{bF}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] FS × TD × Wk ($P > 0.98$); Wk ($P < 0.001$).

[3] FS × Wk ($P < 0.001$); SEM 12.4 g/head/d.

[4] SID CP intake calculated as (total feed intake × calculated dietary SID CP content)/ (pigs per pen × d on feed).

[5] FS × Wk ($P < 0.001$); SEM 0.69 g/head/d.

[6] SID Lys intake calculated as (total feed intake × calculated dietary SID Lys content)/(pigs per pen × d on feed).

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[A,B,C,D,E,F,G] Within a column, means without a common superscript differ ($P \leq 0.05$).

Table A.10. Interactive least squares means of standardized ileal digestible (SID) CP and Lys intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
SID CP Intake[3,4], g/head/d			
8 to 11 wk	236.3^{A}	234.4^{A}	235.7^{A}
11 to 13 wk	291.6^{B}	291.6^{B}	290.8^{B}
13 to 15 wk	337.5^{D}	343.8^{C}	343.9^{C}
15 to 17 wk	377.2^{C}	365.4^{C}	366.5^{DX}
17 to 19 wk	457.1^{aE}	410.5^{bxD}	385.1^{byD}
19 to 21 wk	456.0^{aE}	477.2^{aE}	401.9^{bEY}
21 to 24 wk	455.4^{aE}	479.5^{abE}	499.6^{bF}
SID Lys Intake[5,6], g/head/d			
8 to 11 wk	13.41^{A}	13.30^{A}	13.37^{A}
11 to 13 wk	16.20^{B}	16.20^{B}	16.14^{B}
13 to 15 wk	18.76^{C}	19.10^{C}	19.09^{C}
15 to 17 wk	21.77^{D}	21.04^{D}	21.12^{DX}
17 to 19 wk	24.72^{aE}	22.17^{bxD}	20.82^{byD}
19 to 21 wk	25.43^{aE}	26.59^{aE}	22.41^{bEY}
21 to 24 wk	25.43^{aE}	26.72^{abE}	27.84^{bF}

[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[2] FS × TD × Wk ($P > 0.98$); Wk ($P < 0.001$).

[3] TD × Wk ($P < 0.001$); SEM 12.2 g/head/d.

[4] SID CP intake calculated as (total feed intake × calculated dietary SID CP content)/d on feed.

[5] TD ×Wk ($P < 0.001$); SEM 0.68 g/head/d.

[6] SID Lys intake calculated as (total feed intake × calculated dietary SID Lys content)/d on feed.

[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.11. Interactive least squares means of body composition, carcass dressing and fat-free lean percentages, lean gain, and lean gain efficiency of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
Final BW[3], kg			
PCon	124.7	124.1	126.5
SD	121.8	122.9	123.3
WD	121.3	122.2	123.9
NCon	118.6	120.9	121.5
HCW[4], kg			
PCon	91.1	89.8	91.4
SD	88.0	88.9	88.0
WD	88.1	87.9	88.6
NCon			
Ultrasound backfat[5,6], cm			
PCon	2.12	2.20[x]	1.96[ay]
SD	2.09	2.01	2.05
WD	2.13	2.10	2.07
NCon	1.97	2.16[ax]	1.94[y]
Ultrasound LM area[5,7], cm^2			
PCon	38.3	38.2	38.5
SD	37.1	37.8	37.9
WD	37.3	37.3	37.0
NCon	36.8	37.2	36.9
Lean gain[8,9], g/head/d			
PCon	0.336	0.319	0.339
SD	0.318	0.332	0.333
WD	0.323	0.309	0.317
NCon	0.294	0.302	0.303
Lean gain efficiency[10,11], kg/kg			
PCon	0.141	0.136	0.145
SD	0.134	0.143	0.147
WD	0.134	0.132	0.137
NCon	0.130	0.126	0.134
Lean gain ME efficiency[12,13], kg/Mcal			

PCon	0.042	0.041	0.043
SD	0.041	0.043	0.044
WD	0.041	0.040	0.042
NCon	0.040	0.039	0.041
Carcass dressing[14,15], %			
PCon	72.5	72.2	72.1
SD	71.8	72.4	71.3
WD	72.6	72.1	71.5
NCon	71.0	71.3	70.9
Carcass fat-free lean[16,17], %			
PCon	48.3	48.9	48.8
SD	48.5	49.3	48.5
WD	48.8	48.8	48.6
NCon	49.0	48.8	49.5

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] $P > 0.05$; SEM = 2.1 kg.

[4] $P > 0.05$; SEM = 1.7 kg.

[5] HCW included as a covariate ($P \leq 0.001$).

[6] $P < 0.05$; SEM = 0.07 cm.

[7] $P > 0.05$; SEM = 1.1 cm^2.

[8] Lean gain per day calculated as (final fat-free lean – initial fat-free lean)/number of d on feed; where final fat-free lean = [(2.620 + (0.401 × HCW, kg) – (3.358 × ultrasound 10^{th} rib backfat depth, cm) + (0.306 × ultrasound 10^{th} rib LM, cm^2) + (0.456 × sex of the pig; barrow=1, gilt=2)] and initial fat-free lean = [(0.922 × initial BW, kg) – 3.65] NPPC (2000).

[9] $P > 0.05$; SEM = 0.015 g/head/d.

[10] Lean gain efficiency calculated as total pen fat-free lean gain /total pen feed intake.

[11]$P > 0.05$; SEM = 0.004 kg/kg.

[12] Lean gain ME efficiency calculated as total pen fat-free lean gain /total pen metabolizable energy intake.

[13] $P > 0.05$; SEM = 0.001 kg/Mcal

[14] Carcass dressing percentage was calculated as (final BW before transport/ HCW) × 100.

[15] $P > 0.05$; SEM = 0.4%.

[16] Carcass fat-free lean percentage = [(final fat-free lean)/(HCW)] × 100 (NPPC, 2000).

[17] $P > 0.05$; SEM = 0.6%.

[a] Means without a common superscript differ ($P > 0.05$).

[x,y] Means without a common superscript differ ($P \leq 0.10$).

Table A.12. Body composition of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

FS[1]	PCon			SD			WD			NCon		
Interval between second Improvest® dose and harvest (TD)[2]	TD9	TD7	TD5	TD9	TD7	TD5	TD9	TD7	TD5	TD9	TD7	TD5
Backfat thickness[3,4], cm												
15 WOA	1.09	NA	NA	1.04	NA	NA	1.00	NA	NA	1.02	NA	NA
17 WOA	1.25^{A}	1.24^{A}	NA	1.19^{AB}	1.17^{AB}	NA	1.17^{AB}	1.14^{B}	NA	1.14^{B}	1.16^{AB}	NA
19 WOA	1.49^{aA}	1.41^{abAX}	1.36^{bA}	1.43^{aAB}	1.32^{bBY}	1.32^{bAB}	1.41^{aAB}	1.30^{bB}	1.30^{bAB}	1.34^{xB}	1.32^{xyBY}	1.24^{yB}
21 WOA	1.81^{aA}	1.74^{aAX}	1.60^{bA}	1.74^{axA}	1.63^{byBY}	1.57^{bA}	1.75^{aA}	1.62^{bB}	1.56^{bAB}	1.59^{aB}	1.63^{aBY}	1.45^{bB}
24 WOA	2.21^{xyA}	2.24^{xX}	2.07^{yA}	2.14^{A}	2.09^{Y}	2.05^{A}	2.17^{A}	2.10^{XY}	2.05^{A}	1.91^{aB}	2.11^{bXY}	1.86^{aB}
LM area[3,5], cm^2												
15 WOA	20.45	NA	NA	19.49	NA	NA	18.56	NA	NA	18.75	NA	NA
17 WOA	25.58^{A}	25.41^{A}	NA	24.02^{AB}	23.89^{AB}	NA	22.94^{B}	22.71^{B}	NA	22.62^{B}	22.41^{B}	NA
19 WOA	30.40^{A}	30.27^{A}	30.37^{A}	28.53^{B}	28.47^{BX}	28.50^{BX}	27.54^{BC}	27.26^{B}	27.40^{B}	26.70^{C}	26.47^{BY}	26.88^{BY}
21 WOA	34.92^{AX}	34.83^{A}	35.00^{AX}	33.04^{BY}	33.08^{ABX}	33.10^{BY}	32.36^{BC}	32.02^{B}	32.24^{B}	30.98^{C}	30.72^{BY}	31.42^{B}
24 WOA	39.14^{A}	39.10^{A}	39.35^{A}	37.54^{AB}	37.72^{ABX}	37.73^{AB}	37.40^{AB}	36.97^{AB}	37.30^{AB}	35.46^{B}	35.16^{BY}	36.24^{B}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[3] WOA = week of age.

[4] FS × TD × wk^2; ($P \leq 0.05$); SEM = 0.05 cm

[5] FS × TD × wk^2; ($P \leq 0.46$); SEM = 1.06 cm^2

[a,b] Within a row of a single FS, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row of a single FS, means without a common superscript differ ($P \leq 0.10$).

[A,B,C] Within a row of a single TD, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a row of a single TD, means without a common superscript differ ($P \leq 0.10$).

NA = Not applicable, measures not collected at these time points because the second dose of Improvest® had not been administered yet. Serum collection began at the time of second Improvest® dose.

Table A.13. Radioimmunoassay analysis of serum growth markers of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

FS[1]	PCon			SD			WD			NCon		
Interval between second Improvest® dose and harvest (TD)[2]	TD9	TD7	TD5	TD9	TD7	TD5	TD9	TD7	TD5	TD9	TD7	TD5
IGF-1[3,4], ng/mL												
15 WOA	169.0^{A}	NA	NA	214.5^{B}	NA	NA	189.5^{AB}	NA	NA	191.5^{AB}	NA	NA
17 WOA	166.7^{A}	193.0^{A}	NA	206.7^{B}	232.2^{B}	NA	185.9^{AB}	209.6^{AB}	NA	188.8^{AB}	210.7^{AB}	NA
19 WOA	163.2^{a}	169.3^{ab}	200.4^{bX}	194.2^{a}	198.5^{ab}	231.2^{bY}	180.3^{a}	180.5^{a}	218.5^{bXY}	184.8^{a}	180.9^{a}	222.9^{bXY}
21 WOA	158.5	155.3	156.7	177.0	170.4	174.6	172.8	158.9	172.5	179.3	158.3	179.1
24 WOA	152.6	151.0	133.6	155.0	147.9	135.1	163.2	144.8	146.6	172.5	143.0	156.0
Leptin[3,5], ng/mL												
15 WOA	2.31	NA	NA	2.42	NA	NA	2.21	NA	NA	1.87	NA	NA
17 WOA	2.87	2.67	NA	2.92	2.69	NA	2.82	2.58	NA	2.36	2.09	NA
19 WOA	3.36^{a}	3.24^{aA}	2.36^{bAB}	3.32	3.11^{AB}	2.82^{A}	3.40	3.17^{A}	2.75^{A}	2.75^{a}	2.45^{abB}	1.85^{bB}
21 WOA	3.78^{AB}	3.83^{A}	2.94^{AB}	3.61^{AB}	3.50^{AB}	3.66^{A}	3.93^{A}	3.79^{A}	3.73^{A}	3.02^{B}	2.75^{B}	2.37^{B}
24 WOA	4.14^{AB}	4.43^{A}	3.24^{BCY}	3.79^{AB}	3.84^{AB}	4.28^{AB}	4.44^{A}	4.44^{A}	4.52^{AX}	3.18^{B}	2.98^{B}	2.58^{C}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.

[3] WOA = week of age.

[4] FS × TD × wk^{2}; (P = 0.17) SEM = 19.3 ng/mL.

[5] FS × TD × wk^{2}; (P < 0.05) SEM = 0.42 ng/mL.

^{a,b} Within a row of a single FS, means without a common superscript differ ($P \leq 0.05$).

^{A,B,C} Within a row of a single TD, means without a common superscript differ ($P \leq 0.05$).

^{X,Y} Within a row of a single TD, means without a common superscript differ ($P \leq 0.10$).

NA = Not applicable, measures not collected at these time points because the second dose of Improvest® had not been administered yet. Serum collection began at the time of second Improvest® dose.

Table A.14. Interactive least squares means of body weight before transportation and prior to harvest, hot carcass weight, lairage shrink, and carcass dressing percentages of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
Final BW before transport[3], kg			
PCon	125.6	126.1	125.9
SD	123.4	123.9	125.7
WD	125.0	120.5	121.1
NCon	119.4	122.4	122.5
Final BW at harvest[4], kg			
PCon	121.9	121.5	121.3
SD	119.1	118.6	120.4
WD	120.2	116.2	115.9
NCon	114.9	117.6	116.3
HCW[5], kg			
PCon	93.0	93.1	93.3
SD	91.2	91.3	91.7
WD	92.5	88.6	88.2
NCon	86.3	88.2	88.2
Lairage shrink[6,7], %			
PCon	3.0	3.6	3.6
SD	3.4	4.4	4.3
WD	3.4	3.5	4.2
NCon	3.7	4.3	5.0
Carcass dressing[8,9], %			
PCon	74.1	73.9	74.1
SD	73.9	73.1	72.9
WD	74.0	73.5	72.9
NCon	72.4	72.1	72.0

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[3] $P > 0.05$; SEM = 3.4 kg.

[4] $P > 0.05$; SEM = 3.2 kg.
[5] $P > 0.05$; SEM = 2.3 kg.
[6] $P > 0.05$; SEM = 0.7%.
[7] Lairage shrink loss percentage calculated as [(final BW before transport - BW before harvest)/ final BW before transport] × 100.
[8] $P > 0.05$; SEM = 0.6%.
[9] Dressing percentage calculated as (final BW before transport/ HCW) × 100.

Table A.15. Interactive least squares means lipid and linoleic acid intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies[1]

	Interval between second Improvest® dose and harvest (TD)[2]		
Trait	TD9	TD7	TD5
Backfat thickness, first rib[3,4], cm			
PCon	3.63	3.61	3.56
SD	3.57	3.68	3.27
WD	3.61	3.80	3.48
NCon	3.47	3.47	3.04
Backfat thickness, last rib[3,5], cm			
PCon	2.21	2.19	2.10
SD	2.08	2.29	2.04
WD	2.32	2.13	2.24
NCon	2.31	2.01	2.09
Backfat thickness, last lumbar[3,6], cm			
PCon	1.70	1.52	1.58
SD	1.54	1.79	1.50
WD	1.64	1.56	1.57
NCon	1.87	1.53	1.42
Backfat thickness, 10th rib[3,7], cm			
PCon	1.82	1.72	1.71
SD	1.79	1.96	1.68
WD	1.83	1.80	1.76
NCon	1.88	1.79	1.54
LM area[3,8], cm			
PCon	43.3	43.7	44.0
SD	41.5	41.0	42.1
WD	42.4	42.1	40.5
NCon	42.1	43.1	42.1

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[3] HCW was used as a covariate ($P \leq 0.01$).

[4] $P > 0.05$; SEM = 0.2 cm.
[5] $P > 0.05$; SEM = 0.1 cm.
[6] $P < 0.10$; SEM = 0.1 cm.
[7] $P > 0.05$; SEM = 0.1 cm.
[8] $P > 0.05$; SEM = 1.3 cm^2

Table A.16. Interactive least squares means of chilled carcass weight, chilling loss percentage, and internal carcass fat of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
Chilled carcass wt[3], kg			
PCon	90.8	89.7	90.3
SD	87.5	84.6	88.4
WD	90.9	83.2	85.4
NCon	84.0	83.5	84.9
Chilling loss[4], %			
PCon	3.19	3.06	3.30
SD	3.09	3.22	2.88
WD	2.97	3.38	2.94
NCon	3.23	3.16	3.25
Internal fat (heart and leaf fat)[5], % chilled side wt			
PCon	2.63	2.25	2.09
SD	2.33	2.42	2.21
WD	2.38	2.67	2.30
NCon	2.64	2.24	2.35

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[3] $P > 0.05$; SEM = 3.0 kg.
[4] $P > 0.05$; SEM = 0.38 %.
[5] $P > 0.05$; SEM = 0.19 %

Table A.17. Interactive least squares means of carcass primal weights, and lean and carcass cutting yields of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies*

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
IMPS, % chilled side wt			
401 Ham[3]			
PCon	28.51	28.26	28.43
SD	27.12	26.46	27.74
WD	27.79	26.12	26.83
NCon	25.87	26.19	27.23
402 Trimmed ham[4]			
PCon	25.58	25.14	25.35
SD	23.99	23.36	24.85
WD	25.10	23.24	23.98
NCon	22.85	23.29	24.40
403 Whole shoulder[5]			
PCon	24.11	23.32	24.01
SD	23.75	22.91	23.79
WD	24.38	21.88	23.32
NCon	22.14	22.38	22.72
405 Picnic shoulder[6]			
PCon	11.13	11.20	11.26
SD	10.90	10.21	11.04
WD	10.93	10.06	11.05
NCon	10.08	10.15	10.75
406 Butt shoulder[7]			
PCon	11.30	11.10	11.11
SD	11.10	10.90	11.36
WD	11.48	10.31	10.58
NCon	10.26	10.65	10.37
Whole loin[8]			
PCon	32.20	31.47	31.75
SD	31.34	29.90	31.72
WD	32.57	29.33	30.17
NCon	30.83	29.96	30.13

410 Loin[9]			
PCon	21.52	20.74	22.34
SD	20.34	19.56	21.30
WD	21.15	19.16	20.41
NCon	20.00	19.59	20.23
414 Canadian Back Loin[10]			
PCon	9.48	9.37	9.62
SD	8.69	8.46	9.38
WD	9.30	8.60	8.63
NCon	8.56	8.45	9.02
408 Belly[11]			
PCon	13.65	13.42	13.62
SD	13.32	12.82	13.06
WD	13.00	12.56	13.05
NCon	12.42	12.05	12.13
Lean cutting yield[12,13], %			
PCon	57.84	57.48	57.90
SD	58.15	57.23	58.60
WD	57.15	57.11	58.49
NCon	56.99	58.30	58.55
Carcass cutting yield[14,15], %			
PCon	69.18	68.74	69.45
SD	69.58	68.60	69.73
WD	67.94	68.51	69.94
NCon	68.12	69.20	69.28

[*] n = 6 observations (pens) per treatment combination.

[1] PCon = pigs fed 0% DDGS, standard corn-soybean meal diet throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS in 4 dietary phases, respectively; WD = pigs fed 40% DDGS in phases 1 - 3 and 0% DDGS in phase 4; NCon = pigs fed 40% DDGS throughout growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age; Second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.

[3] $P > 0.05$; SEM = 0.92 %.

[4] $P > 0.05$; SEM = 0.79 %.

[5] $P > 0.05$; SEM = 0.91 %.

[6] $P > 0.05$; SEM = 0.78 %.

[7] $P > 0.05$; SEM = 1.21 %.

[8] $P > 0.05$; SEM = 1.28 %.

[9] $P > 0.05$; SEM = 2.62 %.

[10] $P > 0.05$; SEM = 0.34 %

[11] $P > 0.05$; SEM = 1.06 %
[12] $P > 0.05$; SEM = 3.08 %
[13] [(picnic shoulder + butt shoulder + loin + ham primal cut wt)/ chilled carcass wt] × 100; (Boler et al., 2012).
[14] $P > 0.05$; SEM = 2.71 %
[15] [(picnic shoulder + butt shoulder + loin + ham + belly primal cut wt)/ chilled carcass wt] × 100; (Boler et al., 2012).

Table A.18. Interactive least squares means of objective and subjective pork loin quality assessment of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
pH 45 min[3]			
PCon	6.09	6.08	6.26
SD	6.14	6.23	6.24
WD	6.24	6.17	6.13
NCon	6.30	6.33	6.22
pH 48h[4,5]			
PCon	5.66	5.66	5.64
SD	5.66	5.72	5.66
WD	5.66	5.64	5.67
NCon	5.73	5.62	5.74
LM L*[6]			
PCon	45.4	45.4	46.7
SD	45.6	45.3	44.7
WD	43.4	45.1	44.0
NCon	44.2	43.9	45.1
LM a*[7]			
PCon	-1.60	-1.57	-1.35
SD	-1.35	-1.73	-2.20
WD	-1.70	-1.75	-1.86
NCon	-1.93	-1.99	-2.12
LM b*[8]			
PCon	5.59	5.51	5.71
SD	5.67	5.55	4.94
WD	5.03	5.33	5.19
NCon	5.28	5.01	5.11
Subjective color[9]			
PCon	2.28	2.63	2.41
SD	2.61	2.46	2.43
WD	2.63	2.35	2.24
NCon	2.42	2.25	2.13
Subjective marbling[5,10]			
PCon	1.47	1.40	1.44

SD	1.23	1.23	1.21
WD	1.31	1.31	1.27
NCon	1.16	1.23	1.24
Subjective firmness[11]			
PCon	2.65	2.56	2.60
SD	2.31	2.26	2.39
WD	2.41	2.28	2.15
NCon	2.39	2.03	2.29
Roast purge loss[12], %			
PCon	1.92	2.00	1.71
SD	1.69	1.67	0.98
WD	2.13	1.55	1.96
NCon	1.81	2.02	2.01
Drip loss[13], %			
PCon	2.25	2.47	1.82
SD	1.74	1.95	1.65
WD	1.70	2.21	2.14
NCon	1.81	1.78	1.71
Cook loss[14,15], %			
PCon	20.4	19.0	17.4
SD	19.0	20.8	18.8
WD	17.2	18.3	19.9
NCon	17.9	17.4	19.2
Shear force[16], kg			
PCon	3.13	3.21	2.94
SD	3.22	3.14	3.08
WD	2.99	3.33	3.33
NCon	3.12	2.98	2.98

[1]PCon = pigs fed 0% DDGS, standard corn-soybean meal diet throughout growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS in 4 dietary phases, respectively; WD = pigs fed 40% DDGS in phases 1 - 3 and 0% DDGS in phase 4; NCon = pigs fed 40% DDGS throughout growing-finishing period.

[2] All pigs received the first dose of Improvest® at 11 wk of age; Second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk, respectively before harvest.

[3] $P > 0.05$; SEM = 0.08.

[4] $P > 0.05$; SEM = 0.05.

[5] For 48 h postmortem pH and subjective marbling, data were analyzed and transformed using an exponential (-3) and inverse transformations, respectively. Reported means have been re-transformed.

[6] $P > 0.05$; SEM = 2.6.
[7] $P > 0.05$; SEM = 0.20.
[8] $P > 0.05$; SEM = 0.70.
[9] $P > 0.05$; SEM = 0.15.
[10] $P > 0.05$; SEM = 0.05.
[11] $P > 0.05$; SEM = 0.15.
[12] $P > 0.05$; SEM = 0.34%.
[13] $P > 0.05$; SEM = 0.36%.
[14] $P < 0.05$; SEM = 1.3%.
[15] For cook loss percentages, applying the Tukey adjustment resulted in no significant LS Mean comparisons.
[16] $P > 0.05$; SEM = 0.2%.

Table A.19. Interactive least squares means of overall lipid and linoleic acid intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies[1]

	Interval between second Improvest® dose and harvest (TD)[2]		
Trait	TD9	TD7	TD5
Lipid intake[3,4], g/head/d			
PCon	51.8^{d}	50.9^{d}	50.0^{d}
SD	86.1^{c}	83.7^{c}	81.8^{ck}
WD	95.1^{cj}	92.4^{c}	90.5^{c}
NCon	119.6^{ab}	125.4^{ax}	118.3^{by}
Linoleic acid intake[5,6], g/head/d			
PCon	28.2^{d}	27.7^{d}	27.2^{d}
SD	46.5^{c}	45.3^{c}	44.3^{cy}
WD	51.3^{cx}	49.9^{c}	48.8^{c}
NCon	64.6^{ab}	67.7^{a}	63.8^{b}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] Ether extract intake calculated as (total feed intake × analyzed dietary ether extract)/(pigs per pen × d on feed).

[5] C18:2n6 intake calculated as (total feed intake × analyzed dietary ether extract × analyzed dietary linoleic acid content)/(pigs per pen × d on feed).

[4] $P < 0.10$; SEM = 5.3 g/head/d.

[6] $P < 0.10$; SEM = 2.7 g/head/d.

[a,b,c,d]Means without a common superscript differ ($P \leq 0.05$).

[x,y] Means without a common superscript differ ($P \leq 0.10$).

[j,k] Means without a common superscript differ ($P \leq 0.10$).

Table A.20. Lipid and linoleic acid intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using dried distillers grains with solubles (DDGS) feeding strategies

	Feeding Strategy (FS)[1,2]			
Trait	PCon	SD	WD	NCon
Lipid intake[3,4], g/head/d				
8 to 11 wk	24.2^{aA}	62.1^{bA}	61.9^{bAX}	62.1^{bA}
11 to 13 wk	36.2^{aB}	69.4^{bA}	86.2^{cC}	86.3^{cB}
13 to 15 wk	42.2^{aBC}	81.8^{bBX}	100.7^{cD}	101.5^{cC}
15 to 17 wk	52.0^{aCD}	82.1^{bBX}	115.6^{cE}	116.1^{cD}
17 to 19 wk	57.5^{aD}	92.8^{bCY}	132.9^{cF}	130.7^{cE}
19 to 21 wk	69.5^{aE}	95.7^{bC}	72.9^{aBY}	169.6^{cF}
21 to 24 wk	74.7^{aE}	103.2^{bC}	78.4^{aBC}	181.5^{cG}
Linoleic acid intake[5,6], g/head/d				
8 to 11 wk	13.34^{aA}	33.44^{bA}	33.35^{bA}	33.45^{bA}
11 to 13 wk	19.97^{aB}	37.82^{bxA}	46.41^{cyC}	46.49^{cB}
13 to 15 wk	23.31^{aBC}	44.59^{bBX}	54.21^{cD}	54.61^{cC}
15 to 17 wk	28.40^{aCD}	44.65^{bBX}	62.61^{cE}	62.87^{cD}
17 to 19 wk	31.44^{aD}	$50.46b^{CY}$	71.95^{cF}	70.56^{cE}
19 to 21 wk	37.43^{aE}	51.32^{bC}	39.30^{aB}	91.81^{cF}
21 to 24 wk	40.28^{aE}	55.22^{bC}	42.24^{aBC}	97.96^{cG}

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2] FS × TD × Wk ($P > 0.91$); Wk ($P < 0.001$).

[3] FS × Wk ($P < 0.001$); SEM 5.55 g/head/d.

[4] Lipid intake calculated as (total feed intake × analyzed dietary ether extract)/(pigs per pen × d on feed).

[5] FS × Wk ($P < 0.001$); SEM 2.83 g/head/d.

[6] Linoleic acid intake calculated as (total feed intake × analyzed dietary ether extract × analyzed dietary linoleic acid content)/(pigs per pen × d on feed).

[a,b,c] Within a row, means without a common superscript differ ($P \leq 0.05$).

[x,y] Within a row, means without a common superscript differ ($P \leq 0.10$).

[A,B,C,D,E,F,G] Within a column, means without a common superscript differ ($P \leq 0.05$).

[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.21. Interactive least squares means lipid and linoleic acid intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
Lipid Intake[3,4], g/head/d			
8 to 11 wk	52.8^{A}	52.4^{A}	52.5^{A}
11 to 13 wk	69.2^{B}	70.0^{B}	69.4^{B}
13 to 15 wk	80.5^{C}	82.1^{C}	82.0^{C}
15 to 17 wk	93.1^{D}	90.7^{C}	90.6^{C}
17 to 19 wk	113.2^{aEY}	101.9^{bD}	95.4^{bD}
19 to 21 wk	104.4^{aFX}	109.6^{aD}	91.8^{bD}
21 to 24 wk	103.7^{aF}	110.2^{abD}	114.5^{bD}
C18:2n6 Intake[5,6], g/head/d			
8 to 11 wk	28.52^{A}	28.30^{A}	28.36^{A}
11 to 13 wk	37.52^{B}	37.91^{B}	37.60^{B}
13 to 15 wk	43.63^{C}	44.50^{CX}	44.41^{CX}
15 to 17 wk	50.49^{D}	49.23^{CY}	49.17^{CY}
17 to 19 wk	61.42^{aF}	55.25^{bD}	51.63^{bD}
19 to 21 wk	56.30^{aE}	59.11^{aD}	49.48^{bD}
21 to 24 wk	55.83^{aE}	59.32^{abD}	61.62^{bE}

[1] All pigs received the first dose of Improvest® at 11 wk of age and the second Improvest® dose was administered at either 15, 17, or 19 wk of age which corresponded to pigs receiving the second dose at 9 (TD9), 7 (TD7), or 5 (TD5) wk before harvest, respectively.
[2] FS × TD × Wk ($P > 0.91$); Wk ($P < 0.001$).
[3] FS × Wk ($P < 0.001$); SEM 5.11 g/head/d.
[4] Lipid intake calculated as (total feed intake × analyzed dietary ether extract)/(pigs per pen × d on feed).
[5] FS × Wk ($P < 0.001$); SEM 2.56 g/head/d.
[6] Linoleic acid intake calculated as (total feed intake × analyzed dietary ether extract × analyzed dietary linoleic acid content)/(pigs per pen × d on feed).
[a,b] Within a row, means without a common superscript differ ($P \leq 0.05$).
[A,B,C,D,E,F] Within a column, means without a common superscript differ ($P \leq 0.05$).
[X,Y] Within a column, means without a common superscript differ ($P \leq 0.10$).

Table A.22. Interactive least squares means lipid and linoleic acid intake of immunologically castrated pigs when increasing the time interval between the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies

	Interval between second Improvest® dose and harvest (TD)[1,2]		
Trait	TD9	TD7	TD5
HCW[4], kg			
PCon	93.0	93.1	93.3
SD	91.2	91.3	91.7
WD	92.5	88.6	88.2
NCon	86.3	88.2	88.2
Belly (IMPS #408)[5], % chilled side wt			
PCon	13.65	13.42	13.62
SD	13.32	12.82	13.06
WD	13.00	12.56	13.05
NCon	12.42	12.05	12.13
Belly thickness[6,7], cm			
PCon	3.35	3.27	3.33
SD	3.37	3.30	3.18
WD	3.33	3.40	3.28
NCon	3.28	3.18	3.04
Belly width[6,8], cm			
PCon	22.8	22.7	22.2
SD	22.6	23.1	24.0
WD	22.6	23.5	22.9
NCon	24.0	24.1	23.2
Belly length[6,9], cm			
PCon	61.8	61.2	62.1
SD	62.3	60.3	61.7
WD	60.0	62.0	62.8
NCon	61.1	60.3	62.5
Flop distance[6,10], cm			
PCon	8.63	9.40	8.03
SD	6.93	8.27	6.66
WD	7.04	6.59	7.32
NCon	5.84	5.94	5.80
Flop angle[11,12,13], °			

PCon	16.8	18.1	15.4
SD	13.9	16.1	12.0
WD	13.9	12.6	13.6
NCon	10.7	10.7	9.5

[4] $P > 0.05$; SEM = 2.3 kg.
[5] $P > 0.05$; SEM = 1.06 %.
[6] HCW was used as a covariate ($P < 0.01$).
[7] $P > 0.05$; SEM = 0.04 cm.
[8] $P > 0.05$; SEM = 0.8 cm.
[9] $P > 0.05$; SEM = 1.6 cm.
[10] $P > 0.05$; SEM = 0.7 cm.
[11] $P > 0.05$; SEM = 1.8 cm.
[12] Belly flop angle calculated as: $\cos^{-1}[0.5(L^2) - D^2]/[0.5(L^2)]$, where L = belly length,cm and d = skin-side down, skin-to-skin belly distance,cm (Whitney et al., 2006).
[13] HCW was not used as a covariate ($P > 0.05$).

Table A.23. Interactive least squares means of objective and subjective adipose coor from jowl, belly, and backfat when immunologically castrated pigs were harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies[1]

	Interval between second Improvest® dose and harvest (TD)[2]		
Trait	TD9	TD7	TD5
Jowl adipose, L*[3,4]			
PCon	75.7	73.1	75.6
SD	75.1	74.7	75.7
WD	75.7	74.3	76.4
NCon	75.5	73.5	75.2
Jowl adipose, a*[3,5]			
PCon	0.30	0.99	0.11
SD	0.25	0.35	0.47
WD	0.63	0.62	0.54
NCon	0.22	0.02	0.03
Jowl adipose, b*[3,6]			
PCon	5.45	6.75	5.59
SD	5.33	6.68	6.99
WD	6.14	6.74	7.09
NCon	6.76	6.18	5.90
Back adipose, L*[3,7]			
PCon	76.3	76.2	76.9
SD	76.4	77.3	75.4
WD	77.4	77.3	75.9
NCon	75.4	75.0	75.3
Back adipose, a*[3,8]			
PCon	0.58	0.62	0.31
SD	0.39	0.02	0.51
WD	0.58	1.03	0.49
NCon	-0.03	0.47	0.06
Back adipose, b*[3,9]			
PCon	6.66	6.86	6.42
SD	6.17	6.06	6.53
WD	6.68	7.20	7.06
NCon	6.86	6.26	6.52
Belly adipose, L*[3,10]			
PCon	78.2	78.2	79.3

SD	77.9	78.6	77.8
WD	78.0	77.0	77.9
NCon	76.7	77.1	75.8
Belly adipose, a*[3,11]			
PCon	0.05	0.20	-0.31
SD	-0.21	0.07	-0.08
WD	0.10	0.13	0.17
NCon	-0.39	0.08	-0.42
Belly adipose, b*[3,12]			
PCon	5.90	5.46	5.01
SD	5.36	5.30	4.46
WD	5.37	5.75	6.23
NCon	4.70	6.01	5.52
JCS,[13,14]			
PCon	1.00	1.06	1.19
SD	1.19	1.19	1.06
WD	1.25	1.25	1.38
NCon	1.19	1.31	1.50

[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2] All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[3] Objective Hunter color values L* (100 = white, 0 = black), a* (positive = more red, negative = more green), b* (positive = more yellow, negative = more blue).
[4] $P > 0.05$; SEM = 1.9.
[5] $P > 0.05$; SEM = 0.34.
[6] $P > 0.05$; SEM = 0.94.
[7] $P > 0.05$; SEM = 1.7.
[8] $P > 0.05$; SEM = 0.41.
[9] $P > 0.05$; SEM = 0.87.
[10] $P > 0.05$; SEM = 0.7.
[11] $P > 0.05$; SEM = 0.26.
[12] $P > 0.05$; SEM = 0.62.
[13] $P > 0.05$; SEM = 0.12.
[14] JCS = Japanese Color Score (1 = white and 4 = yellow).

Table A.24. Comprehensive fatty acid composition and calculated iodine vale (IV) of jowl, back, and belly fat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies (FS)[1]

	Jowl				Back				Belly					*P* value
Item	PCon	SD	WD	NCon	PCon	SD	WD	NCon	PCon	SD	WD	NCon	SEM	FS × Depot
Crude Fat[2], %	88.67	88.44	88.63	87.97	87.99	88.9	88.44	88.33	88.44	88.32	87.85	88.74		0.04
SFA														
C8:0 – Caprylic	0.0112	0.0121	0.0118	0.0121	0.0124	0.0121	0.0119	0.0121	0.0125	0.0122	0.0119	0.0114	0.0004	0.20
C10:0 – Capric	0.093^{ax}	0.082^{bc}	0.074^{defl}	0.068^{f}	0.087^{by}	0.080^{cdm}	0.079^{cde}	0.069^{f}	0.093^{ax}	0.085^{bc}	0.080^{czk}	0.073^{efn}	0.002	0.01
C12:0 – Lauric	0.079	0.074	0.069	0.066	0.082	0.078	0.077	0.070	0.081	0.077	0.073	0.069	0.002	0.30
C14:0 – Myristic	1.40	1.28	1.22	1.17	1.42	1.33	1.31	1.20	1.45	1.33	1.28	1.21	0.02	0.37
C15:0 – Pentadecylic	0.066	0.067	0.071	0.077	0.059	0.06	0.057	0.072	0.063	0.062	0.064	0.072	0.004	0.18
C16:0 – Palmitic	23.52^{c}	22.08^{ey}	21.73^{ef}	20.43^{g}	25.93^{a}	24.59^{b}	25.18^{ab}	22.44^{de}	24.46^{b}	23.23^{cd}	23.01^{cdx}	21.11^{fg}	0.28	< 0.01
C18:0 – Stearic	10.52^{cd}	9.76^{de}	9.7^{de}	8.62^{f}	14.18^{a}	12.97^{b}	13.75^{ab}	10.98^{c}	11.11^{c}	10.52^{cd}	10.42^{cd}	8.94^{ef}	0.26	< 0.01
C20:0 – Arachidic	0.234	0.233	0.227	0.220	0.274	0.268	0.270	0.262	0.250	0.247	0.250	0.238	0.007	0.99
C21:0 – Heneicosylic	0.043	0.044	0.046	0.059	0.036	0.037	0.035	0.048	0.041	0.048	0.050	0.062	0.004	0.57
C22:0 – Behenoic	0.119^{bcd}	0.142^{abk}	0.125^{bcdy}	0.136^{abcm}	0.121^{bcd}	0.121^{bcd}	0.111^{cdl}	0.128^{abcd}	0.106^{dn}	0.134^{abcd}	0.136^{abcm}	0.154^{ax}	0.012	0.02
C23:0 – Tricosylic	0.022	0.021	0.023	0.022	0.023	0.021	0.022	0.021	0.020	0.020	0.020	0.023	0.001	0.85
C24:0 – Lignoceric	0.048	0.042	0.041	0.037	0.044	0.046	0.039	0.038	0.03	0.034	0.026	0.028	0.003	0.51
MUFA														
C14:1n-9 - Myristoleic	0.026^{ab}	0.022^{bc}	0.022^{bc}	0.023^{bc}	0.023^{bc}	0.021^{c}	0.021^{c}	0.023^{bc}	0.029^{a}	0.023^{bc}	0.024^{bc}	0.021^{c}	0.001	< 0.01
C15:1n-10 - Pentadecenoic	0.0017	0.0003	0.0006	0.0012	0.0010	0.0006	0.0007	0.0015	0.0003	0.0003	0.0001	0.0007	0.0005	0.83
C18:1n9t - Elaidic	0.184	0.179	0.181	0.172	0.170	0.153	0.161	0.153	0.168	0.171	0.171	0.149	0.006	0.53
C18:1n9 - Oleic	38.97^{b}	37.23^{cd}	37.03^{cd}	36.11^{dey}	37.38^{cx}	35.42^{e}	35.45^{e}	32.89^{f}	40.19^{a}	37.83^{bc}	37.43^{cx}	35.38^{e}	0.53	< 0.01
C20:1n9 - Gonodic	0.92^{ax}	0.86^{abc}	0.84^{bcy}	0.85^{abc}	0.84^{bc}	0.85^{abc}	0.80^{c}	0.83^{bc}	0.86^{abc}	0.85^{abc}	0.89^{ab}	0.85^{abc}	0.02	0.02
C24:1n9 - Nervonic	0.132	0.161	0.164	0.177	0.113	0.161	0.134	0.163	0.124	0.136	0.152	0.171	0.006	0.32
PUFA														
C18:2n6 - Linoleic	13.18^{f}	18.05^{bc}	19.44^{b}	23.38^{a}	10.64^{g}	15.67^{de}	14.73^{ef}	22.67^{a}	11.38^{g}	16.32^{de}	17.06^{cd}	22.87^{a}	0.91	< 0.01

C18:3n3 - Linolenic	0.73^{cdex}	0.78^{bcd}	0.81^{b}	0.91^{a}	0.60^{g}	0.64^{fgy}	0.62^{g}	0.80^{bc}	0.60^{g}	0.64^{fg}	0.71^{def}	0.79^{bcd}	0.04	0.04
C20:2 - Eicosadienoic	0.74^{f}	0.89^{bcde}	0.91^{bc}	1.12^{a}	0.69^{f}	0.77^{f}	0.71^{f}	0.98^{b}	0.78^{efy}	0.80^{cdef}	0.90^{bcdx}	0.98^{b}	0.04	< 0.001
C20:3n3 - Homo-α-linolenic	0.128	0.144	0.155	0.162	0.070	0.094	0.078	0.109	0.066	0.075	0.079	0.076	0.008	0.11
C20:3n6 - Homo-γ-linolenic	0.016	0.013	0.015	0.017	0.036	0.037	0.044	0.050	0.010	0.010	0.010	0.016	0.009	0.91
C20:4n6 - Arachidonic	0.397^{cde}	0.452^{abcx}	0.469^{ab}	0.496^{a}	0.335^{e}	0.372^{de}	0.337^{e}	0.364^{de}	0.380^{de}	0.386^{dey}	0.376^{de}	0.425^{bcd}	0.019	0.06
C20:5n3 - Eicosapentaenoic	0.003	0.005	0.002	0.004	0.013	0.010	0.011	0.008	0.012	0.015	0.010	0.012	0.002	0.21
C22:2n6 - Docosadienoic	0.006	0.005	0.007	0.006	< 0.0001	< 0.0001	0.0005	< 0.0001	0.0104	0.0133	0.0070	0.0103	0.0025	0.19
C22:4n6 - Adrenic	0.001^{d}	0.003^{d}	0.003^{d}	0.003^{d}	0.004^{cdy}	0.007^{bcd}	0.006^{bcd}	0.012^{abx}	0.012^{abx}	0.016^{a}	0.005^{bcd}	0.012^{abc}	0.002	< 0.01
C22:5n3 - Clupanodonic	ND	ND	ND	ND	0.0020	ND	0.0027	ND	0.0013	0.0022	ND	ND	0.0010	0.22
C22:6n3 - Docosahexaenoic	0.038	0.034	0.039	0.038	0.056	0.058	0.054	0.042	0.030	0.030	0.026	0.024	0.005	0.50
Total SFA[3]	36.6^{cd}	34.2^{ef}	33.8^{fg}	31.3^{h}	42.7^{a}	40.0^{b}	41.3^{ab}	35.7^{de}	38.1^{c}	36.2^{d}	35.8^{de}	32.4^{gh}	0.5	< 0.01
Total MUFA[4]	45.3^{ab}	42.9^{bcd}	43.5^{bcd}	42.2^{cd}	43.0^{bcd}	41.6^{d}	41.7^{dy}	38.6^{e}	47.5^{a}	44.3^{bcx}	44.1^{bcx}	41.4^{d}	0.7	0.07
Total PUFA[5]	15.3^{gm}	20.4^{c}	21.9^{c}	26.2^{ax}	12.1^{h}	17.2^{efkl}	16.2^{fg}	24.5^{by}	13.3^{h}	18.4^{de}	19.2^{dj}	25.3^{ab}	1.0	< 0.01
Total n3[6]	0.90^{cd}	0.97^{bc}	1.01^{b}	1.12^{a}	0.74^{ef}	0.80^{dex}	0.76^{ef}	0.96^{bc}	0.71^{fy}	0.76^{ef}	0.83^{de}	0.90^{cd}	0.05	0.05
Total n6[7]	13.6^{f}	18.5^{bc}	19.9^{b}	23.9^{a}	11.0^{g}	16.1^{de}	15.1^{ef}	23.1^{a}	11.8^{g}	16.8^{de}	17.5^{cd}	23.3^{a}	0.9	< 0.01
n6:n3	15.6^{fg}	19.3^{eyjl}	19.8^{de}	21.4^{cdx}	15.2^{gl}	20.3^{de}	20.2^{de}	24.3^{b}	17.2^{fk}	22.6^{bc}	21.4^{cdx}	26.6^{a}	0.5	< 0.01
IV-AOCS[8]	61.9^{f}	68.5^{cd}	70.6^{bc}	76.7^{a}	55.3^{g}	62.1^{f}	60.5^{f}	72.1^{b}	59.4^{f}	65.5^{e}	66.7^{de}	74.9^{a}	1.3	< 0.01
IV-Meadus[8]	64.6^{f}	71.5^{cd}	73.8^{bc}	80.3^{a}	57.8^{g}	64.9^{f}	63.1^{f}	75.2^{b}	62.1^{f}	68.3^{e}	69.6^{de}	78.0^{a}	1.3	< 0.001
IV difference[9]	2.68^{d}	3.05^{bc}	3.16^{b}	3.59^{a}	2.57^{d}	2.81^{cd}	2.63^{d}	3.08^{bc}	2.69^{d}	2.77^{cd}	2.81^{cd}	3.14^{b}	0.11	< 0.01

[1]PCon = pigs fed corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in 4 dietary phases, respectively; WD = pigs fed 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = pigs fed 40% DDGS diets throughout the growing-finishing period.

[2] For crude fat, with Tukey adjustment Diet × Depot comparisons were not significantly different ($P > 0.05$).

[3] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.

[4] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10 Margaroleic] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].

[5] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2 Eicosadienoic] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].

[6]Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].

[7] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].
[8]Iodine Value (IV)-AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV- Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930) (Meadus et al., 2010).
[a,b,c,d,e,f,g] Within a row, means without a common superscript differ ($P < 0.05$).
[9] IV differences = (IV-AOCS) – (IV-Meadus).
[x,y] Within a row, means without a common superscript differ ($P < 0.10$).
[j,k] Within a row, means without a common superscript differ ($P < 0.10$).
[l,m] Within a row, means without a common superscript differ ($P < 0.10$).

Table A.25. Main effects of fatty acid composition of jowl, belly, and backfat from immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest dose when using corn dried distillers grains with solubles (DDGS) feeding strategies

	Feeding strategy (FS)[1]					Interval between 2nd Improvest dose and harvest (TD)[2]				Fat Depot				*P* value[3,4,5]		
Item	PCon	SD	WD	NCon	SEM	TD9	TD7	TD5	SEM	Jowl	Back	Belly	SEM	FS	TD	Depot
Crude Fat, %	88.36	88.55	88.31	88.35	0.18	88.59	88.31	88.27	0.15	88.43	88.34	88.41	0.14	0.78	0.28	0.88
SFA																
C8:0 – Caprylic	0.012	0.0121	0.0118	0.0118	0.0002	0.0120	0.0122	0.0118	0.0002	0.0118	0.0121	0.0120	0.0002	0.74	0.34	0.40
C10:0 – Capric	0.091^{a}	0.082^{b}	0.078^{c}	0.07^{d}	0.002	0.079	0.081	0.081	0.002	0.079^{a}	0.079^{a}	0.083^{b}	0.002	< 0.01	0.18	< 0.01
C12:0 – Lauric	0.081^{a}	0.076^{bx}	0.073^{by}	0.069^{c}	0.002	0.074	0.075	0.074	0.002	0.072^{a}	0.077^{bx}	0.075^{by}	0.002	< 0.01	0.80	< 0.01
C14:0 – Myristic	1.422^{a}	1.31^{b}	1.27^{b}	1.19^{c}	0.02	1.31	1.31	1.28	0.02	1.27^{a}	1.32^{b}	1.32b	0.02	< 0.01	0.19	< 0.01
C15:0 - Pentadecylic	0.062^{a}	0.063^{a}	0.064^{a}	0.073^{b}	0.003	0.064	0.065	0.069	0.003	0.070^{a}	0.062^{b}	0.065^{c}	0.003	< 0.01	0.13	< 0.01
C16:0 – Palmitic	24.64^{a}	23.3^{b}	23.31^{b}	21.32^{c}	0.24	23.25^{a}	23.30^{ax}	22.88^{y}	0.23	21.94^{a}	24.54^{b}	22.95^{c}	0.21	< 0.01	0.05	< 0.01
C18:0 – Stearic	11.93^{a}	11.08^{b}	11.29^{b}	9.51^{c}	0.20	11.10	11.00	10.76	0.19	9.65^{a}	12.97^{b}	10.25^{c}	0.17	< 0.01	0.19	< 0.01
C20:0 – Arachidic	0.253	0.249	0.249	0.240	0.005	0.249	0.248	0.246	0.004	0.228^{a}	0.269^{b}	0.246^{c}	0.004	0.23	0.91	< 0.01
C21:0 - Heneicosylic	0.040^{a}	0.043^{a}	0.044^{a}	0.056^{b}	0.003	0.047	0.045	0.045	0.003	0.048	0.039	0.050	0.003	< 0.01	0.61	< 0.01
C22:0 – Behenoic	0.116^{a}	0.132^{bc}	0.124^{ab}	0.139^{c}	0.010	0.127^{ab}	0.121^{a}	0.136^{b}	0.010	0.130^{a}	0.121^{b}	0.133^{a}	0.010	< 0.01	0.01	0.03
C23:0 - Tricosylic	0.021	0.021	0.021	0.022	0.001	0.022	0.021	0.021	0.001	0.022^{a}	0.022^{a}	0.020^{b}	0.001	0.47	0.20	0.01
C24:0 - Lignoceric	0.041^{a}	0.040^{ax}	0.036^{b}	0.034^{by}	0.002	0.036	0.04	0.037	0.002	0.042^{a}	0.042^{a}	0.029^{b}	0.002	0.01	0.13	< 0.01
MUFA																
C14:1n-9 - Myristoleic	0.026^{a}	0.022^{b}	0.022^{b}	0.023^{b}	0.001	0.023	0.023	0.024	0.001	0.023^{ab}	0.022^{a}	0.024^{b}	0.001	< 0.01	0.15	< 0.01
C15:1n-10 – Pentadecenoic	0.0010	0.0004	0.0004	0.0011	0.0003	0.0007	0.0050	0.0010	0.0003	0.0010^{ax}	0.0010^{ax}	0.0003^{y}	0.0002	0.25	0.51	0.07
C18:1n9t - Elaidic	0.174^{a}	0.167^{ab}	0.171^{a}	0.158^{b}	0.004	0.166	0.170	0.168	0.004	0.179^{a}	0.159^{b}	0.164^{b}	0.004	< 0.01	0.69	< 0.01
C18:1n9 - Oleic	38.85^{a}	36.83^{b}	36.64^{b}	34.8^{c}	0.49	36.9	36.94	36.49	0.48	37.34^{ax}	35.29^{b}	37.71^{ay}	0.46	< 0.01	0.17	< 0.01
C20:1n9 - Gonodic	0.87	0.85	0.84	0.84	0.02	0.85	0.86	0.85	0.01	0.87^{a}	0.83^{b}	0.86^{a}	0.01	0.13	0.73	< 0.01
C24:1n9 - Nervonic	0.123^{a}	0.153^{b}	0.150^{b}	0.170^{c}	0.004	0.148	0.147	0.152	0.004	0.158^{ax}	0.137^{b}	0.152^{ay}	0.004	< 0.01	0.33	< 0.01
PUFA																
C18:2n6 - Linoelic	11.73^{a}	16.68^{b}	17.08^{b}	22.97^{c}	0.86	16.7^{a}	16.81^{a}	17.83^{b}	0.84	18.51^{a}	15.92^{c}	16.91^{b}	0.83	< 0.01	< 0.01	< 0.01

C18:3n3 - Linoleic	0.64[a]	0.69[b]	0.71[b]	0.83c	0.04	0.71[a]	0.70[a]	0.75[b]	0.04	0.81[a]	0.66[b]	0.68[b]	0.04	< 0.01	< 0.01	< 0.01
C20:2 - Eicosadienoic	0.74[a]	0.82[b]	0.84[b]	1.02[c]	0.03	0.86	0.85	0.86	0.03	0.91[a]	0.79[c]	0.86[b]	0.03	< 0.01	0.75	< 0.01
C20:3n3 - Homo-α-linolenic	0.089[a]	0.105[b]	0.104[b]	0.116[b]	0.006	0.100	0.104	0.105	0.006	0.147[a]	0.088[b]	0.074[c]	0.006	< 0.01	0.59	< 0.01
C20:3n6 - Homo-γ-linolenic	0.021	0.020	0.023	0.028	0.008	0.025	0.020	0.023	0.008	0.021[a]	0.020[b]	0.023[a]	0.008	0.29	0.35	< 0.01
C20:4n6 - Arachidonic	0.371[a]	0.403[bc]	0.394[ab]	0.428[c]	0.015	0.388[x]	0.398	0.412[y]	0.014	0.453[a]	0.352[c]	0.392[b]	0.014	< 0.01	0.08	< 0.01
C20:5n3 - Eicosapentaenoic	0.009	0.010	0.008	0.008	0.001	0.009	0.008	0.009	0.001	0.004	0.010	0.012	0.001	0.37	0.73	< 0.01
C22:2n6 - Docosadienoic	0.0055	0.0063	0.0048	0.0056	0.0023	0.0050	0.0055	0.0062	0.0022	0.0062[b]	0.0103[a]	0.0002[c]	0.0022	0.68	0.52	< 0.01
C22:4n6 - Adrenic	0.0057[ab]	0.0086[acx]	0.0048[by]	0.0090[c]	0.0013	0.0055[a]	0.0063[ax]	0.0093[by]	0.0011	0.0024[a]	0.0073[b]	0.0114[c]	0.0011	0.01	< 0.01	< 0.01
C22:5n3 - Clupanodonic	0.0011	0.0007	0.0009	ND	0.0006	ND	0.0009	0.0011	0.0006	ND	0.0012	0.0009	0.0005	0.59	0.27	0.18
C22:6n3 - Docosahexaenoic	0.042	0.041	0.039	0.035	0.004	0.039	0.04	0.039	0.004	0.037[b]	0.052[a]	0.028c	0.004	0.32	0.94	< 0.01
Total SFA[6]	39.1[a]	36.8[b]	36.9[b]	33.1[c]	0.4	36.8[x]	36.7	36.0[y]	0.4	34.0[a]	39.9[c]	35.6[b]	0.4	< 0.01	0.07	< 0.01
Total MUFA[7]	45.3[a]	42.9[b]	43.1[b]	40.8[c]	0.6	42.9	43.2	43.0	0.6	43.5[ax]	41.2[b]	44.3[ay]	0.5	< 0.01	0.73	< 0.01
Total PUFA[8]	13.6[a]	18.7[b]	19.1[b]	25.3[c]	0.9	18.8[a]	18.8[a]	19.9[b]	0.9	21.0[a]	17.5[c]	19.0[b]	0.9	< 0.01	< 0.01	< 0.01
Total n3[9]	0.78[a]	0.84[b]	0.87[b]	0.99[c]	0.05	0.86[a]	0.85[a]	0.90[b]	0.05	1.00[a]	0.82[b]	0.80[b]	0.05	< 0.01	0.02	< 0.01
Total n6[10]	12.1[a]	17.1[b]	17.5[b]	23.4[c]	0.9	17.1[a]	17.2[a]	18.3[b]	0.9	19.0[a]	16.3[c]	17.3[b]	0.8	< 0.01	< 0.01	< 0.01
n6:n3	16.0	20.7	20.5	24.1	0.3	20.2	20.4	20.5	0.3	19.0	21.9	20.0	0.2	< 0.01	0.67	< 0.01
IV-AOCS[11]	58.9[a]	65.4[b]	65.9[b]	74.6[c]	1.2	65.5[a]	65.8[a]	67.2[b]	1.2	69.4[a]	66.6[b]	62.5[c]	1.2	< 0.01	< 0.01	< 0.01
IV-Meadus[11]	61.5[a]	68.2[b]	68.8[b]	77.8[c]	1.3	68.4[a]	68.7[a]	70.2[b]	1.3	72.5[a]	69.5[b]	65.2[c]	1.3	< 0.01	< 0.01	< 0.01
IV difference[12]	2.65[a]	2.88[b]	2.86[b]	3.27[c]	0.09	2.89	2.89	2.97	0.09	3.12[a]	2.77[b]	2.85[b]	0.09	< 0.01	0.26	< 0.01

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] FS × TD × depot; ($P > 0.05$) except for C18:1n-9 - Oleic, C18:3n-3 - linolenic, Total n3, and n3:n6 (See Appendix, Figure A.2, Chapter 6, Figure 6.3, Appendix Figures A.6, A.7, respectively).

[4]TD × depot; ($P > 0.05$) except for C23:0 Tricosylic, C15:1n-10 Pentadecenoic, C20:3n3 - Homo-α-linolenic, C20:3n6 - Homo-γ-linolenic, C22:5n3 - Clupanodonic, n3:n6 (See Appendix Figures A.1, A.3, A.5, A.5, A.5, A.8, respectively).

[5]FS × TD; ($P > 0.05$) except for C10:0 Capric, C14:1n-9 - Myristoleic, C18:1n9t - Elaidic, C20:3n3 - Homo-α-linolenic, C20:3n6 - Homo-γ-linolenic (See Appendix Figures A.1, A.3, A.3, A.4, A.4, respectively).

[6] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.

[7] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].

[8] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].
[9] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].
[10] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].
[11] Iodine Value (IV) - AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV- Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930; Meadus et al., 2010).
[12] IV differences = (IV-AOCS) – (IV-Meadus).
[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).
[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

Table A.26. Interactive least squares means of fatty acid composition and caluclated iodine value (IV) from jowl, belly, and backfat of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and fed corn dried distillers grains with solubles (DDGS) feeding strategies

Interval between 2nd Improvest® dose and harvest (TD)[1]	TD9				TD7				TD5					*P* value
Feeding Strategy (FS)[2]	PCon	SD	WD	NCon	PCon	SD	WD	NCon	PCon	SD	WD	NCon	SEM	FS × TD
Item														
Crude Fat, %	88.86	88.17	88.65	88.70	87.95	88.93	88.21	88.15	88.28	88.56	88.06	88.19	0.31	0.21
SFA														
C8:0 – Caprylic	0.0118	0.0125	0.0118	0.0117	0.0122	0.0124	0.0120	0.0120	0.0120	0.0116	0.0117	0.0118	0.0004	0.85
C10:0 – Capric	0.089^{abx}	0.079^{cd}	0.080^{cdy}	0.067^{fw}	0.094^{a}	0.084^{bc}	0.076^{cdez}	0.069^{ef}	0.090^{ab}	0.084^{bc}	0.077^{cde}	0.074^{def}	0.002	0.05
C12:0 – Lauric	0.079	0.077	0.074	0.068	0.083	0.077	0.073	0.067	0.080	0.075	0.072	0.070	0.002	0.39
C14:0 – Myristic	1.41	1.34	1.28	1.19	1.45	1.32	1.28	1.19	1.40	1.28	1.25	1.20	0.03	0.63
C15:0 – Pentadecylic	0.060	0.062	0.057	0.076	0.059	0.060	0.066	0.073	0.068	0.068	0.069	0.072	0.005	0.41
C16:0 – Palmitic	24.72	23.57	23.29	21.43	24.93	23.67	23.24	21.35	24.26	22.67	23.39	21.19	0.32	0.36
C18:0 – Stearic	11.91	11.46	11.17	9.87	12.16	11.22	11.31	9.32	11.73	10.57	11.39	9.34	0.31	0.38
C20:0 – Arachidic	0.257	0.248	0.259	0.231	0.250	0.262	0.237	0.244	0.252	0.238	0.251	0.245	0.008	0.10
C21:0 – Heneicosylic	0.044	0.045	0.045	0.054	0.037	0.042	0.042	0.058	0.040	0.042	0.044	0.056	0.004	0.74
C22:0 – Behenoic	0.122	0.126	0.123	0.136	0.107	0.125	0.122	0.132	0.118	0.147	0.127	0.150	0.011	0.53
C23:0 – Tricosylic	0.024	0.021	0.022	0.022	0.019	0.020	0.022	0.022	0.021	0.021	0.021	0.021	0.001	0.17
C24:0 – Lignoceric	0.040	0.039	0.031	0.033	0.042	0.045	0.039	0.034	0.039	0.038	0.037	0.036	0.003	0.73
MUFA														
C14:1n-9 - Myristoleic	0.027^{ax}	0.022^{abcy}	0.021^{bc}	0.020^{c}	0.026^{a}	0.022^{abcy}	0.021^{bc}	0.024^{abc}	0.025^{ab}	0.023^{abc}	0.025^{ab}	0.024^{abc}	0.001	0.01
C15:1n-10 - Pentadecenoic	0.0004	0.0003	0.0003	0.0021	0.0007	0.0005	0.0003	0.0006	0.0020	0.0003	0.0009	0.0007	0.0005	0.20
C18:1n9t - Elaidic	0.175	0.171	0.166	0.152	0.172	0.162	0.172	0.173	0.176^{x}	0.170	0.175	0.150^{y}	0.006	0.10
C18:1n9 - Oleic	39.01	36.63	37.21	34.77	38.77	37.14	36.63	35.20	38.77	36.72	36.08	34.42	0.57	0.56
C20:1n9 - Gonodic	0.88	0.88	0.82	0.83	0.86	0.86	0.84	0.87	0.87	0.83	0.85	0.84	0.02	0.25
C24:1n9 - Nervonic	0.122	0.146	0.150	0.175	0.119	0.152	0.145	0.170	0.129	0.160	0.155	0.166	0.007	0.50
PUFA														
C18:2n6 - Linoleic	11.40	16.41	16.46	22.55	11.64	15.87	17.15	22.58	12.16	17.76	17.63	23.79	0.96	0.84
C18:3n3 - Linolenic	0.64	0.68	0.69	0.82	0.62	0.65	0.72	0.82	0.66	0.73	0.74	0.86	0.05	0.84
C20:2 - Eicosadienoic	0.80	0.80	0.83	1.01	0.70	0.81	0.83	1.04	0.71	0.83	0.85	1.03	0.04	0.29
C20:3n3 - Homo-α-linolenic	0.088^{b}	0.117^{ab}	0.098^{ab}	0.097^{ab}	0.084^{b}	0.101^{ab}	0.108^{ab}	0.124^{a}	0.092^{b}	0.096^{ab}	0.106^{ab}	0.126^{a}	0.008	0.02
C20:3n6 - Homo-γ-linolenic	0.014^{a}	0.018^{ab}	0.029^{ab}	0.040^{b}	0.019^{ab}	0.023^{ab}	0.020^{ab}	0.019^{ab}	0.029^{ab}	0.019^{ab}	0.020^{ab}	0.024^{ab}	0.009	0.06
C20:4n6 - Arachidonic	0.364	0.413	0.382	0.392	0.368	0.390	0.403	0.431	0.380	0.409	0.396	0.462	0.019	0.20
C20:5n3 - Eicosapentaenoic	0.009	0.008	0.008	0.010	0.009	0.010	0.006	0.010	0.010	0.011	0.008	0.007	0.002	0.54
C22:2n6 - Docosadienoic	0.006	0.005	0.004	0.005	0.006	0.005	0.005	0.006	0.006	0.008	0.005	0.006	0.003	0.84

C22:4n6 - Adrenic	0.004	0.005	0.003	0.009	0.007	0.006	0.005	0.007	0.006	0.015	0.006	0.010	0.002	0.11
C22:5n3 - Clupanodonic	ND	ND	ND	ND	ND	0.0018	0.0006	ND	0.0020	0.0018	0.0021	ND	0.001	0.80
C22:6n3 - Docosahexaenoic	0.045	0.041	0.036	0.033	0.042	0.046	0.040	0.032	0.038	0.035	0.042	0.040	0.005	0.32
Total SFA[3]	39.2	39.6	38.6	33.6	39.6	37.3	36.9	33.0	38.6	35.6	37.1	32.8	0.6	0.30
Total MUFA[4]	45.2	42.0	43.6	40.6	44.9	42.0	43.0	41.3	45.8	43.1	42.5	40.4	0.8	0.47
Total PUFA[5]	13.3	18.4	18.5	24.8	13.4	17.8	19.2	24.9	14.0	19.8	19.7	26.2	1.0	0.85
Total n3[6]	0.78	0.85	0.83	0.96	0.76	0.85	0.87	0.98	0.80	0.87	0.90	1.04	0.05	0.72
Total n6[7]	11.8	16.8	16.9	23.0	12.0	16.3	17.6	23.0	12.6	18.2	18.1	24.3	1.0	0.83
n6:n3	15.4	20.1	20.6	24.5	16.6	20.5	20.4	24.1	16.1	21.6	20.4	23.8	0.5	0.29
IV-AOCS[8]	58.5	64.6	65.3	73.6	58.5	64.2	66.0	74.3	59.6	67.2	66.5	75.7	1.4	0.81
IV-Meadus[8]	61.2	67.5	68.1	76.7	61.1	67.1	68.9	77.6	62.2	70.1	69.4	79.1	1.5	0.81
IV difference[9]	2.71	2.86	2.82	3.16	2.59	2.86	2.87	3.25	2.64	2.92	2.91	3.40	0.11	0.56

[1]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[2] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[3] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.

[4] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10 Margaroleic] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].

[5] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2 Eicosadienoic] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].

[6] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].

[7] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].

[8] Iodine Value (IV) - AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV – Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930; Meadus et al., 2010).

[9] IV differences = (IV-AOCS) – (IV-Meadus).

[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).

[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

Table A.27. Interactive least squares means of fatty acid composition and calculated iodine value (IV) of jowl, belly, and backfat of immunologically castrated pigs harvested at 5, 7, or 9 wk after the second Improvest® dose and harvest while using corn dried distillers grains with solubles (DDGS) feeding strategies[1]

Fat depot	Jowl			Back			Belly				*P* value
Interval between 2nd Improvest® dose and harvest (TD)[2]	TD9	TD7	TD5	TD9	TD7	TD5	TD9	TD7	TD5	SEM	TD × Depot
Crude Fat, %	88.65	88.22	88.42	88.64	88.44	88.16	88.49	88.28	88.23	0.25	0.90
SFA											
C8:0 – Caprylic	0.0119	0.0120	0.0114	0.0126	0.0126	0.0120	0.0121	0.0118	0.0120	0.0003	0.39
C10:0 – Capric	0.077	0.080	0.080	0.078	0.079	0.080	0.081	0.083	0.084	0.002	0.96
C12:0 – Lauric	0.072	0.073	0.072	0.077	0.077	0.077	0.075	0.076	0.075	0.002	1.00
C14:0 – Myristic	1.27	1.27	1.25	1.32	1.33	1.30	1.32	1.33	1.29	0.02	0.94
C15:0 – Pentadecylic	0.068	0.070	0.073	0.061	0.060	0.065	0.062	0.064	0.069	0.003	0.74
C16:0 – Palmitic	22.08	22.02	21.72	24.59	24.74	24.28	23.09	23.13	22.63	0.26	0.93
C18:0 – Stearic	9.78	9.61	9.55	13.12	13.14	12.64	10.40	10.26	10.08	0.24	0.79
C20:0 – Arachidic	0.228	0.229	0.228	0.272	0.270	0.264	0.246	0.246	0.247	0.006	0.92
C21:0 – Heneicosylic	0.048	0.049	0.046	0.042	0.037	0.038	0.051	0.048	0.052	0.004	0.36
C22:0 – Behenoic	0.13	0.12	0.15	0.12	0.12	0.12	0.13	0.13	0.14	0.01	0.16
C23:0 – Tricosylic	0.023^{x}	0.021	0.022	0.023^{x}	0.023	0.020	0.020	0.019^{y}	0.021	0.001	0.06
C24:0 – Lignoceric	0.041	0.044	0.042	0.041	0.046	0.039	0.026	0.031	0.031	0.003	0.65
MUFA											
C14:1n-9 - Myristoleic	0.0226	0.0233	0.0244	0.0212	0.0228	0.0227	0.0238	0.0248	0.0246	0.0009	0.93
C15:1n-10 - Pentadecenoic	0.0014	0.0008	0.0007	0.0007	0.0004	0.0018	ND	0.0004	0.0005	0.0004	0.09
C18:1n9t - Elaidic	0.184	0.175	0.179	0.150	0.163	0.164	0.164	0.170	0.161	0.006	0.14
C18:1n9 - Oleic	37.49	37.42	37.11	35.14	35.70	35.03	38.08	37.69	37.35	0.51	0.25
C20:1n9 - Gonodic	0.88	0.87	0.85	0.82	0.84	0.82	0.86	0.86	0.87	0.02	0.58
C24:1n9 - Nervonic	0.160	0.157	0.158	0.135	0.133	0.142	0.148	0.150	0.158	0.005	0.51
PUFA											
C18:2n6 - Linoleic	18.08	18.51	18.94	15.69	15.17	16.91	16.33	16.75	17.65	0.88	0.24
C18:3n3 - Linolenic	0.80	0.80	0.83	0.65	0.64	0.70	0.67	0.66	0.72	0.04	0.78
C20:2 - Eicosadienoic	0.92	0.89	0.93	0.76	0.81	0.78	0.90	0.84	0.86	0.04	0.14
C20:3n3 - Homo-α-linolenic	0.137^{a}	0.155^{a}	0.150^{a}	0.083^{bc}	0.093^{b}	0.088^{bc}	0.081^{bc}	0.064^{b}	0.077^{bc}	0.007	0.05
C20:3n6 - Homo-γ-linolenic	0.019^{bc}	0.009^{c}	0.018^{bc}	0.048^{a}	0.034^{ab}	0.044^{a}	0.009^{c}	0.016^{bc}	0.008^{c}	0.009	0.09
C20:4n6 - Arachidonic	0.440	0.452	0.468	0.330	0.358	0.368	0.393	0.383	0.399	0.018	0.62
C20:5n3 - Eicosapentaenoic	0.004	0.004	0.003	0.012	0.009	0.011	0.012	0.012	0.013	0.001	0.65
C22:2n6 - Docosadienoic	0.0060	0.0075	0.0052	ND	0.0004	0.0001	0.0090	0.0085	0.0133	0.0024	0.06
C22:4n6 - Adrenic	0.0013	0.0035	0.0022	0.0061	0.0044	0.0115	0.0090	0.0109	0.0143	0.0017	0.12
C22:5n3 - Clupanodonic	ND	ND	ND	ND	0.0005	0.0030	ND	0.0023	0.0003	0.0008	0.04
C22:6n3 - Docosahexaenoic	0.037	0.036	0.038	0.054	0.057	0.045	0.024	0.026	0.033	0.005	0.11
Total SFA[3]	34.24	34.01	33.68	40.14	40.29	39.32	35.91	35.79	35.12	0.46	0.89

Total MUFA[4]	42.62	44.11	43.72	41.31	41.26	41.11	44.65	44.30	44.02	0.67	0.26
Total PUFA[5]	20.51	20.93	21.44	17.26	16.76	18.50	18.48	18.80	19.82	0.92	0.37
Total n3[6]	0.98	1.00	1.02	0.80	0.80	0.85	0.79	0.76	0.84	0.05	0.52
Total n6[7]	18.55	18.98	19.43	16.08	15.57	17.33	16.75	17.16	18.08	0.89	0.25
n6:n3	19.03^{c}	19.00^{c}	19.11^{c}	20.22^{bc}	19.31^{c}	20.50^{bc}	21.22^{ab}	22.78^{a}	21.84^{ab}	0.41	0.01
IV-AOCS[8]	68.8	69.5	70.0	61.8	61.5	64.0	66.0	66.3	67.7	1.3	0.44
IV-Meadus[8]	71.8	72.6	73.2	64.5	64.4	66.9	68.8	69.0	70.6	1.4	0.54
IV difference[9]	3.09	3.07	3.19	2.69	2.81	2.81	2.88	2.79	2.89	0.10	0.42

[1] FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] Total SFA calculated as [C8:0] + [C10:0] + [C12:0], [C14:0] + [C15:0] + [C16:0] + [C18:0] + [C20:0] + [C21:0] + [C22:0] + [C23:0] + [C24:0]. Fatty acids C6:0, C11:0, C13:0, and C17:0 were not detectable in these adipose samples.

[4] Total MUFA calculated as [C14:1n-9 Myristoleic] + [C16:1n-6 Palmitoleic] + [C17:1n-10 Margaroleic] + [C18:1n-9t Elaidic] + [C18:1n-9 Oleic] + [C18:1n-11 Vaccenic] + [C20:1n-9 Gonodic] + [C22:1n-9 Erucic] + [C24:1n-9 Nervonic].

[5] Total PUFA calculated as [C18:2t Linoelaidic] + [C18:2n-6 Linoleic] + [C18:3n-3 Linolenic] + [C20:2 Eicosadienoic] + [C20:2n-3 homo-γ-linolenic] + [C20:4n-6 Arachidonic] + [C22:2n-6 Docosadienoic] + [C20:5n-3 Eicosapentaenoic (EPA)] + [C22:4n-6 Adrenic] + [C22:5n-3Clupanodonic] + [C22:6n-3 Docosahexaenoic (DHA)].

[6] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].

[7] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].

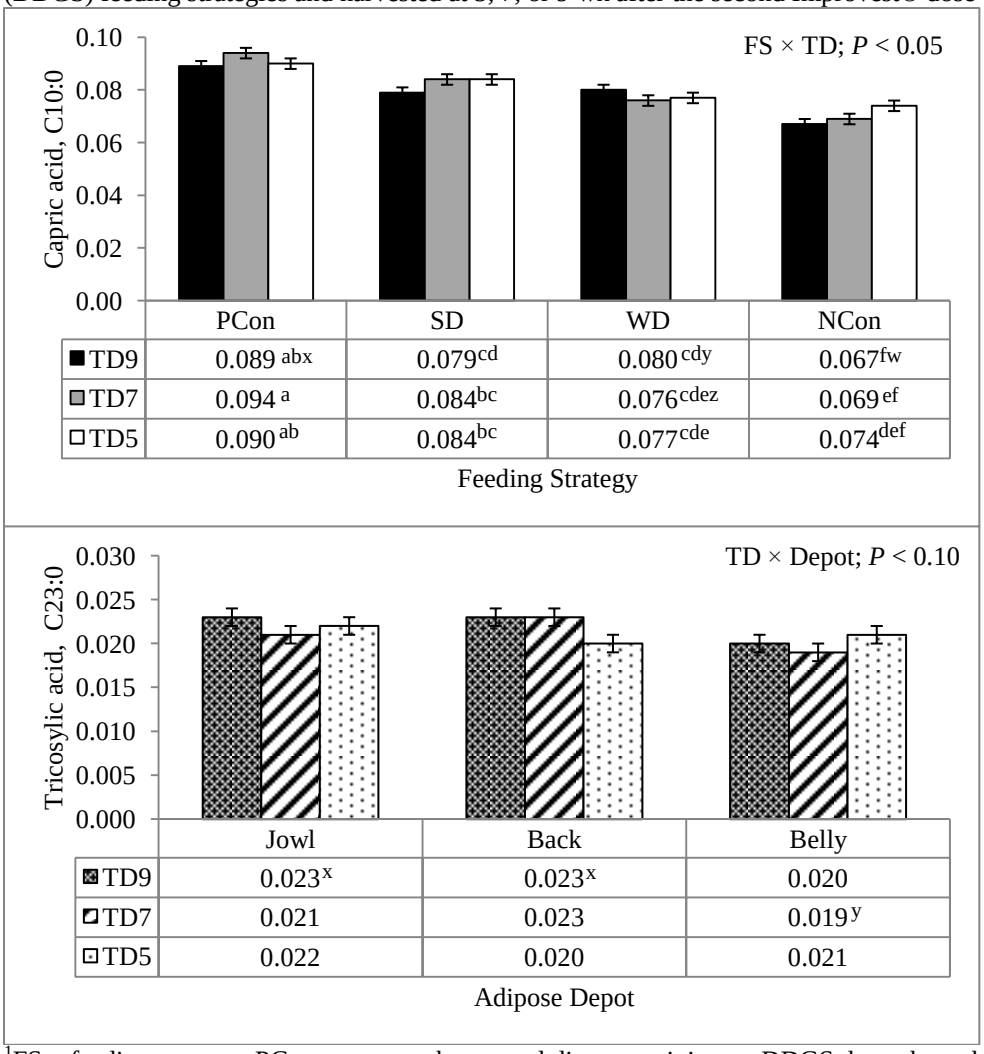
[8] Iodine Value (IV) - AOCS = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.785) + [C22:1] × 0.723) (AOCS, 1998); IV - Meadus = ([C16:1] × 0.95) + ([C18:1] × 0.86) + ([C18:2] × 1.732) + ([C18:3] × 2.616) + ([C20:1] × 0.795) + ([C20:2] × 1.570) + ([C20:3] × 2.380) + ([C20:4] × 3.190) + ([C20:5] × 4.010) + ([C22:4] × 2.930) + ([C22:5] × 3.680) + ([C22:6] × 2.930; Meadus et al., 2010).

[9] IV differences = (IV-AOCS) – (IV-Meadus).

[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).

[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

Figure A.1. Interactive least square means of saturated fatty acids (capric acid and tricosylic acid) of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose[1,2]



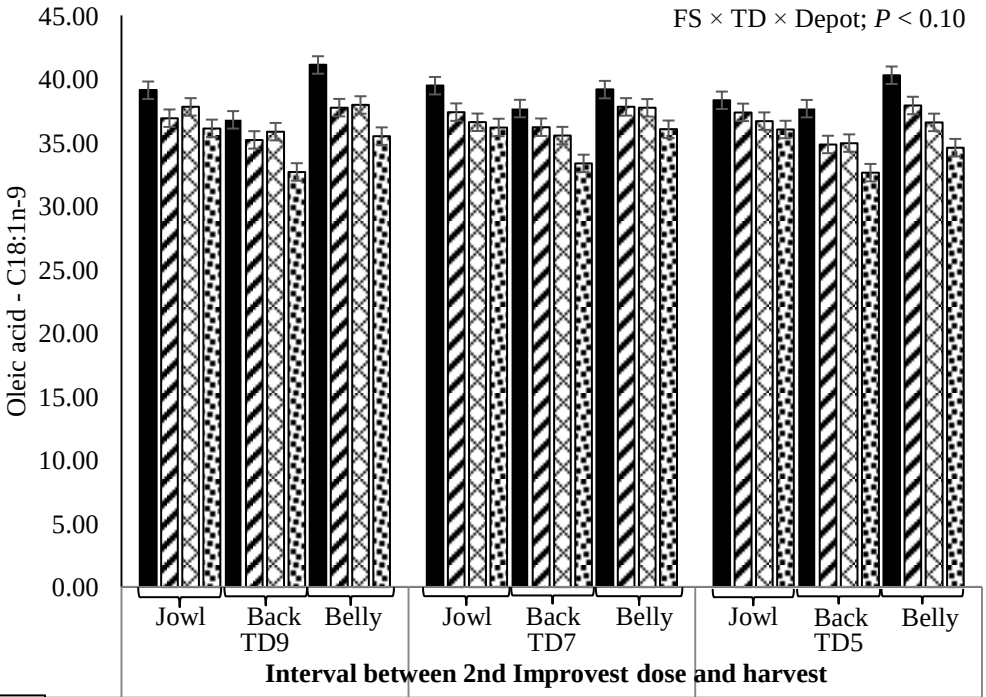
[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[a,b,c,d,e,f] Least squares means with different superscripts differ ($P \leq 0.05$).

[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

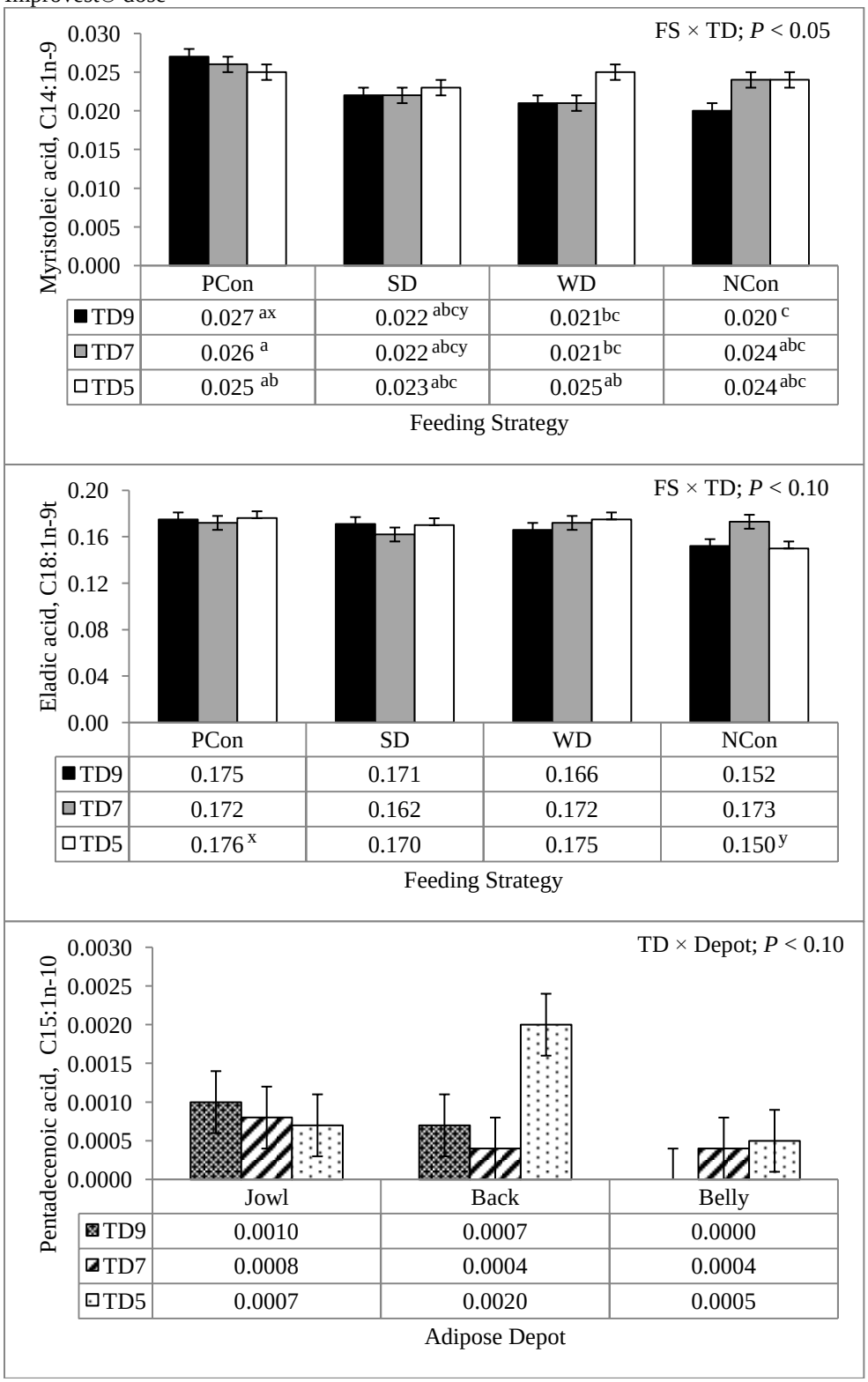
Figure A.2. Interactive least square means of oleic acid content of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose[1,2]



		TD9	TD7	TD5
Jowl	PCon	39.12	39.48	38.33
	SD	36.92	37.40	37.38
	WD	37.81	36.60	36.69
	NCon	36.12	36.19	36.03
Back	PCon	36.79	37.68	37.67
	SD	35.21	36.21	34.85
	WD	35.86	35.55	34.96
	NCon	32.69	33.36	32.63
Belly	PCon	41.11	39.17	40.30
	SD	37.75	37.81	37.92
	WD	37.96	37.74	36.59
	NCon	35.51	36.05	34.60

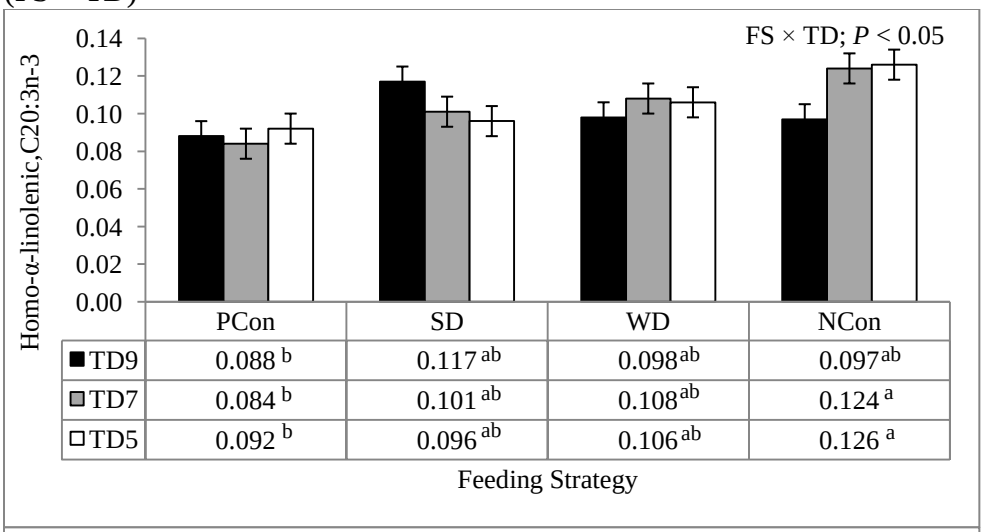
[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

Figure A.3. Interactive least square means of monounsaturated fatty acids (myristoleic acid, eladic acid, and pentadecenoic acid) of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose[1,2]



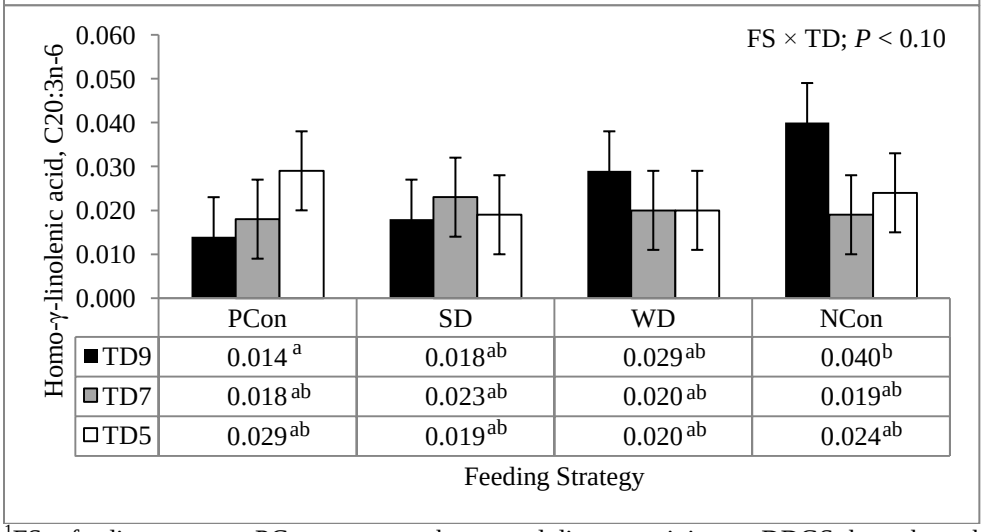
[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).
[x,y] Least squares means with different superscripts differ ($P \leq 0.10$).

Figure A.4. Interactive least square means of polyunsaturated fatty acids (homo-γ-linolenic acid and homo-α-linolenic acid) of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies (FS) and harvested at 5, 7, or 9 wk after the second Improvest® dose (FS × TD)[1,2]



	PCon	SD	WD	NCon
TD9	0.088 b	0.117 ab	0.098 ab	0.097 ab
TD7	0.084 b	0.101 ab	0.108 ab	0.124 a
TD5	0.092 b	0.096 ab	0.106 ab	0.126 a

Feeding Strategy



	PCon	SD	WD	NCon
TD9	0.014 a	0.018 ab	0.029 ab	0.040 b
TD7	0.018 ab	0.023 ab	0.020 ab	0.019 ab
TD5	0.029 ab	0.019 ab	0.020 ab	0.024 ab

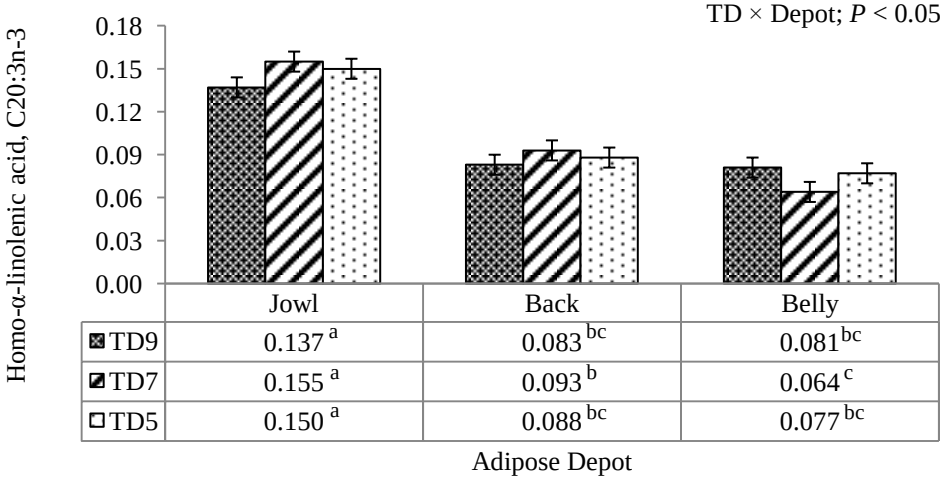
Feeding Strategy

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

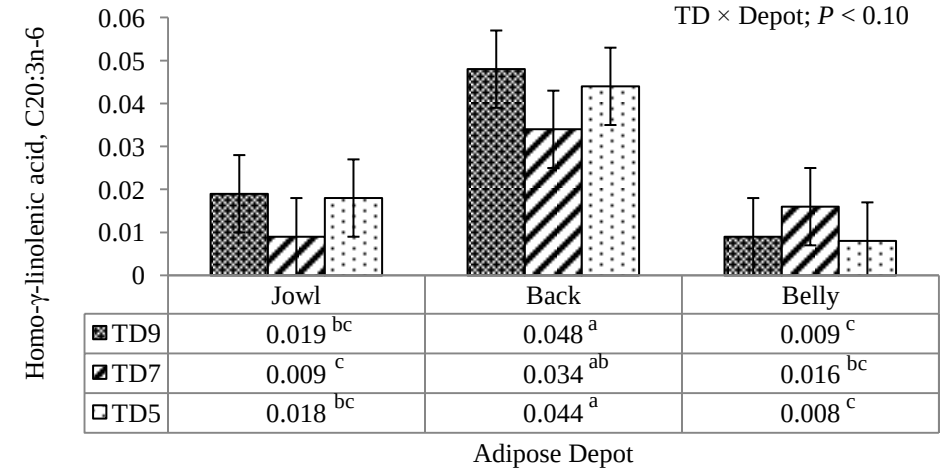
[a,b] Least squares means with different superscripts differ ($P \leq 0.05$).

Figure A.5. Interactive least square means of polyunsaturated fatty acids (homo-γ-linolenic acid, homo-α-linolenic acid, and clupanodonic acid) of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies (FS) and harvested at 5, 7, or 9 wk after the second Improvest® dose (TD × Depot)[1,2]



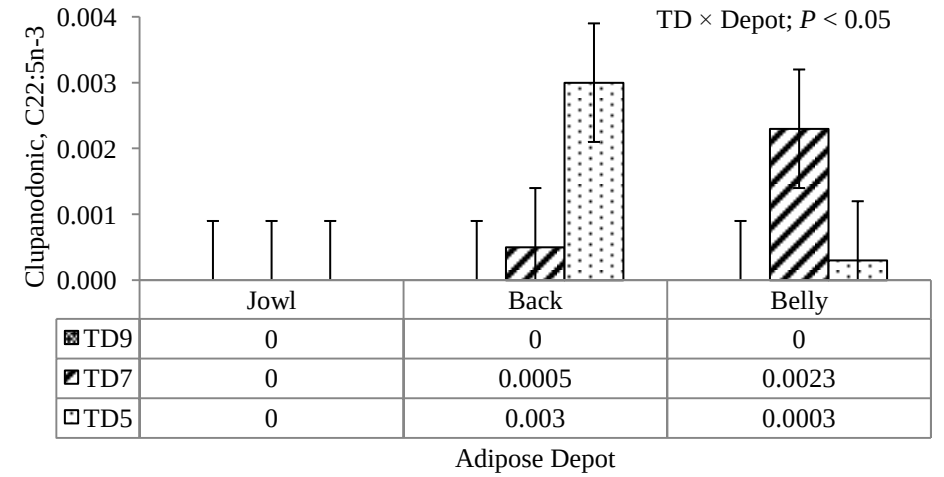
	Jowl	Back	Belly
TD9	0.137 [a]	0.083 [bc]	0.081 [bc]
TD7	0.155 [a]	0.093 [b]	0.064 [c]
TD5	0.150 [a]	0.088 [bc]	0.077 [bc]

Adipose Depot



	Jowl	Back	Belly
TD9	0.019 [bc]	0.048 [a]	0.009 [c]
TD7	0.009 [c]	0.034 [ab]	0.016 [bc]
TD5	0.018 [bc]	0.044 [a]	0.008 [c]

Adipose Depot

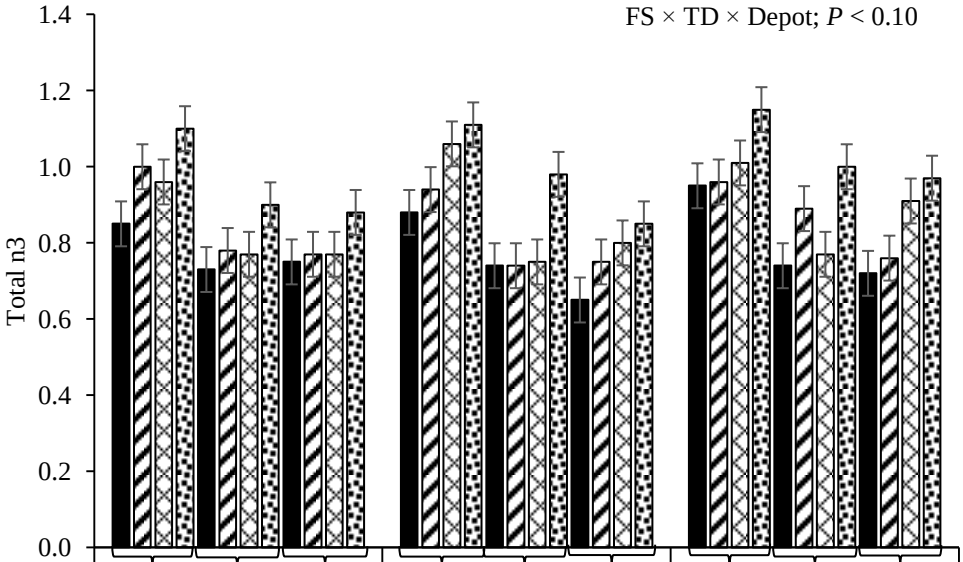


	Jowl	Back	Belly
TD9	0	0	0
TD7	0	0.0005	0.0023
TD5	0	0.003	0.0003

Adipose Depot

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.
[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).

Figure A.6. Interactive least square means of total n3 fatty acid content of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose[1,2,3]



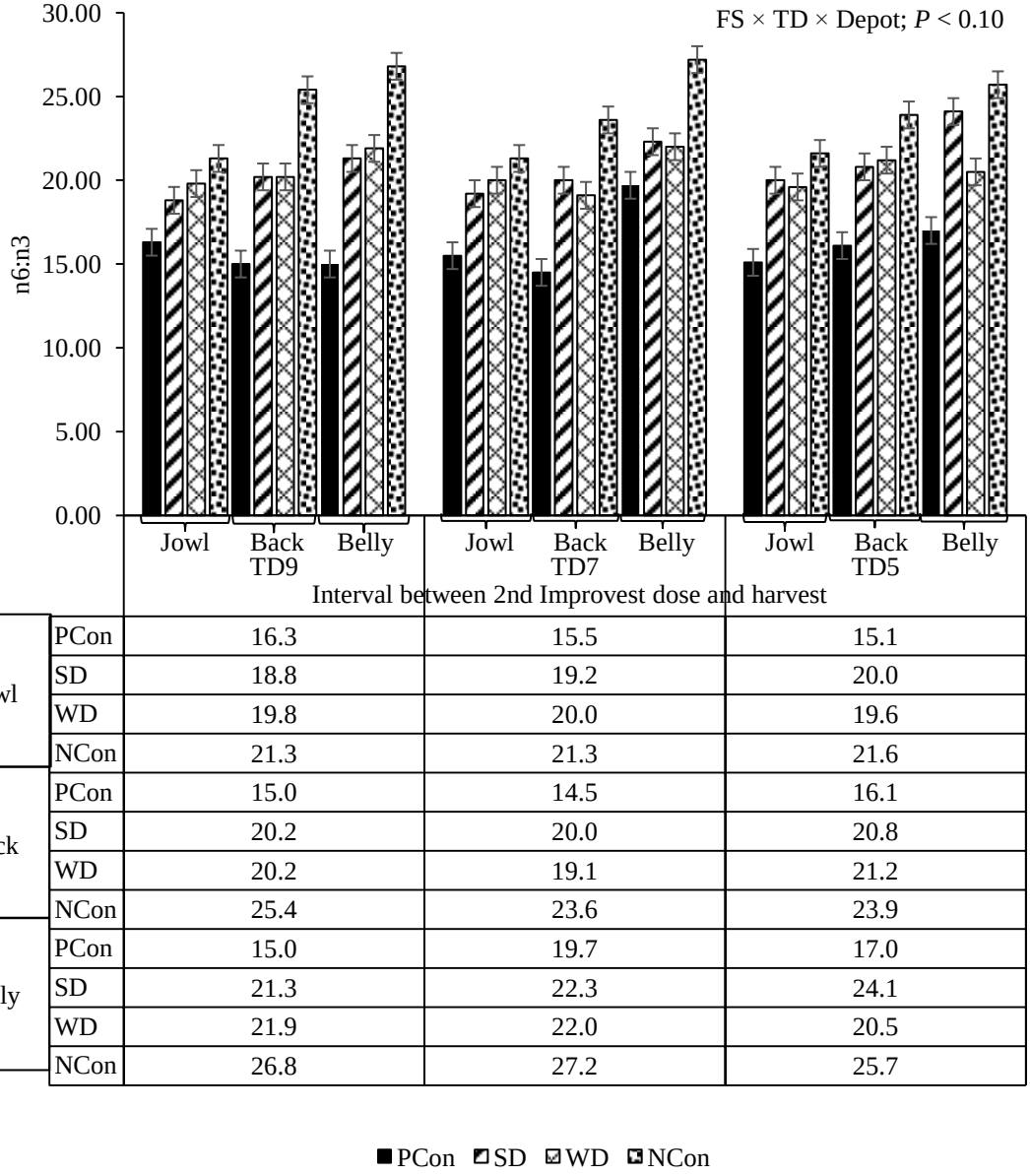
Jowl	PCon	0.85	0.88	0.95
	SD	1.00	0.94	0.96
	WD	0.96	1.06	1.01
	NCon	1.10	1.11	1.15
Back	PCon	0.73	0.74	0.74
	SD	0.78	0.74	0.89
	WD	0.77	0.75	0.77
	NCon	0.90	0.98	1.00
Belly	PCon	0.75	0.65	0.72
	SD	0.77	0.75	0.76
	WD	0.77	0.80	0.91
	NCon	0.88	0.85	0.97

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3]Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].

Figure A.7. Interactive least square means of n6:n3 fatty acid ratio of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose[1,2,3,4]

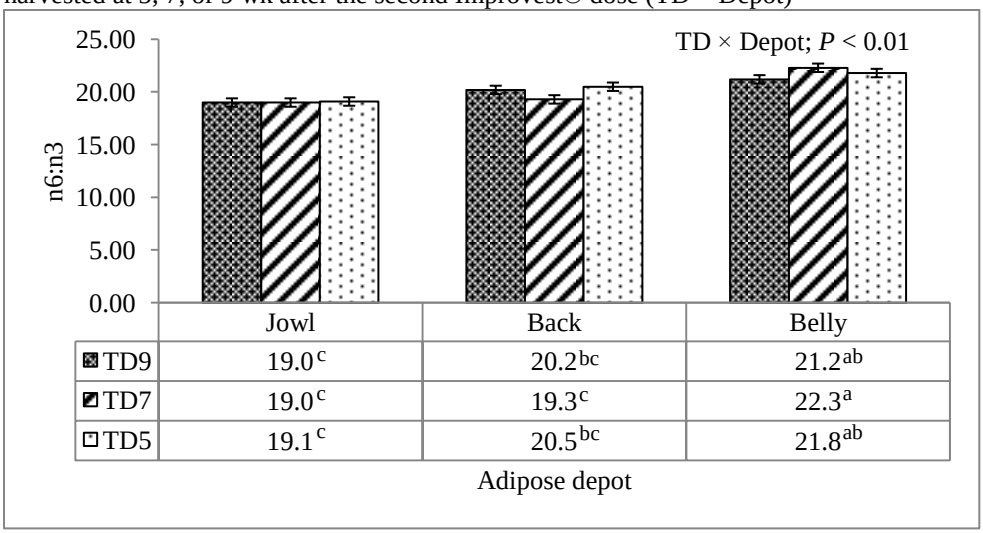


		TD9	TD7	TD5
Jowl	PCon	16.3	15.5	15.1
	SD	18.8	19.2	20.0
	WD	19.8	20.0	19.6
	NCon	21.3	21.3	21.6
Back	PCon	15.0	14.5	16.1
	SD	20.2	20.0	20.8
	WD	20.2	19.1	21.2
	NCon	25.4	23.6	23.9
Belly	PCon	15.0	19.7	17.0
	SD	21.3	22.3	24.1
	WD	21.9	22.0	20.5
	NCon	26.8	27.2	25.7

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.
[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[3] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].
[4] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].

Figure A.8. Interactive least square means of n6:n3 fatty acid ratio of jowl, belly, and backfat from immunologically castrated pigs fed corn dried distillers grains with solubles (DDGS) feeding strategies and harvested at 5, 7, or 9 wk after the second Improvest® dose (TD × Depot)[1,2,3,4]



	Jowl	Back	Belly
TD9	19.0^{c}	20.2^{bc}	21.2^{ab}
TD7	19.0^{c}	19.3^{c}	22.3^{a}
TD5	19.1^{c}	20.5^{bc}	21.8^{ab}

Adipose depot

[1]FS = feeding strategy; PCon = corn-soybean meal diets containing no DDGS throughout the growing-finishing period; SD = pigs fed 40, 30, 20, and 10% DDGS diets in phase 1 to 4 , respectively; WD = 40% DDGS diets in phases 1 to 3 and 0% DDGS diet in phase 4; NCon = 40% DDGS diets throughout the growing-finishing period.

[2]All pigs received the first dose of Improvest® at 11 wk of age; second Improvest® dose was administered at either 15, 17, or 19 wk of age, corresponding to pigs receiving the second dose at 9, 7, or 5 wk, respectively before harvest.

[a,b,c] Least squares means with different superscripts differ ($P \leq 0.05$).

[3] Total n3 fatty acids calculated as [C18:3n-3] + [C20:2n-3] + [C20:5n-3] + [C22:5n-3] + [C22:6n-3].

[4] Total n6 fatty acids calculated as [C18:2n-6] + [C20:3n-6] + [C20:4n-6] + [C22:2n-6] + [C22:4n-6].